



INDIAN AGRICULTURAL
RESEARCH INSTITUTE, NEW DELHI

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CARNEGIE INSTITUTION OF WASHINGTON

YEAR BOOK No. 47

July 1, 1947—June 30, 1948

With Administrative Reports through December 10, 1948



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CARNEGIE INSTITUTION OF WASHINGTON
WASHINGTON, D. C.

1948

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PRESIDENT AND TRUSTEES

PRESIDENT

VANNEVAR BUSH

BOARD OF TRUSTEES

WALTER S. GIFFORD, *Chairman*
ELIHU ROOT, JR., *Vice-Chairman*
LEWIS H. WEED, *Secretary*

JAMES F. BELL
ROBERT WOODS BLISS
LINDSAY BRADFORD
FREDERIC A. DELANO
HOMER L. FERGUSON
W. CAMERON FORBES
JAMES FORRESTAL
WALTER S. GIFFORD
HERBERT HOOVER

FRANK B. JEWETT
ERNEST O. LAWRENCE
ALFRED L. LOOMIS
ROBERT A. LOVETT
ROSWELL MILLER
HENRY S. MORGAN
SEELEY G. MUDD
WILLIAM I. MYERS
HENNING W. PRENTIS, JR.

*GORDON S. RENTSCHLER
ELIHU ROOT, JR.
HENRY R. SHEPLEY
†RICHARD P. STRONG
CHARLES P. TAFT
JUAN T. TRIPPE
JAMES W. WADSWORTH
‡FREDERIC C. WALCOTT
LEWIS H. WEED

Executive Committee

ROBERT WOODS BLISS
VANNEVAR BUSH

WALTER S. GIFFORD, *Chairman*
FRANK B. JEWETT
HENRY S. MORGAN
ELIHU ROOT, JR.

HENRY R. SHEPLEY
LEWIS H. WEED

Finance Committee

ALFRED L. LOOMIS

LINDSAY BRADFORD, *Chairman*
HENRY S. MORGAN
HENNING W. PRENTIS, JR.

ELIHU ROOT, JR.

Auditing Committee

HOMER L. FERGUSON

FREDERIC A. DELANO, *Chairman*

JAMES W. WADSWORTH

Nominating Committee

JAMES F. BELL

WALTER S. GIFFORD
HENRY S. MORGAN

HENNING W. PRENTIS, JR.

Committee on Astronomy

ROSWELL MILLER

SEELEY G. MUDD, *Chairman*
ELIHU ROOT, JR.

JUAN T. TRIPPE

Committee on Terrestrial Sciences

HOMER L. FERGUSON

FRANK B. JEWETT, *Chairman*

ERNEST O. LAWRENCE

Committee on Biological Sciences

ALFRED L. LOOMIS

LEWIS H. WEED, *Chairman*
WILLIAM I. MYERS
HENNING W. PRENTIS, JR.

CHARLES P. TAFT

Committee on Historical Research

JAMES F. BELL

HENRY R. SHEPLEY, *Chairman*
ROBERT WOODS BLISS

JAMES W. WADSWORTH

* Deceased March 3, 1948.

† Deceased July 4, 1948.

‡ Resigned May 20, 1948.

FORMER PRESIDENTS AND TRUSTEES

PRESIDENTS

DANIEL COIT GILMAN, 1902-1904 ROBERT SIMPSON WOODWARD, 1904-1920
 JOHN CAMPBELL MERRIAM, *President* 1921-1938; *President Emeritus* 1939-1945

TRUSTEES

ALEXANDER AGASSIZ	1904-05	ANDREW J. MELLON	1924-37
GEORGE J. BALDWIN	1925-27	DARIUS O. MILLS	1902-09
THOMAS BARBOUR	1934-46	S. WEIR MITCHELL	1902-14
JOHN S. BILLINGS	1902-13	ANDREW J. MONTAGUE	1907-35
ROBERT S. BROOKINGS	1910-29	WILLIAM W. MORROW	1902-29
JOHN L. CADWALADER	1903-14	WILLIAM CHURCH OSBORN	1927-34
WILLIAM W. CAMPBELL	1929-38	JAMES PARMELEE	1917-31
JOHN J. CARTY	1916-32	WM. BARCLAY PARSONS	1907-32
WHITEFOORD R. COLE	1925-34	STEWART PATON	1916-42
CLEVELAND H. DODGE	1903-23	GEORGE W. PEPPER	1914-19
WILLIAM E. DODGE	1902-03	JOHN J. PERSHING	1930-43
CHARLES P. FENNER	1914-24	HENRY S. PRITCHETT	1906-36
SIMON FLEXNER	1910-14	GORDON S. RENTSCHLER	1946-48
WILLIAM N. FREW	1902-15	ELIHU ROOT	1902-37
LYMAN J. GAGE	1902-12	JULIUS ROSENWALD	1929-31
CASS GILBERT	1924-34	MARTIN A. RYERSON	1908-28
FREDERICK H. GILLETT	1924-35	THEOBALD SMITH	1914-34
DANIEL C. GILMAN	1902-08	JOHN C. SPOONER	1902-07
JOHN HAY	1902-05	WILLIAM BENSON STOREY	1924-39
MYRON T. HERRICK	1915-29	RICHARD P. STRONG	1934-48
ABRAM S. HEWITT	1902-03	WILLIAM H. TAFT	1906-15
HENRY L. HIGGINSON	1902-19	WILLIAM S. THAYER	1929-32
ETHAN A. HITCHCOCK	1902-09	CHARLES D. WALCOTT	1902-27
HENRY HITCHCOCK	1902-02	FREDERIC C. WALCOTT	1931-48
WILLIAM WIRT HOWE	1903-09	HENRY P. WALCOTT	1910-24
CHARLES L. HUTCHINSON	1902-04	WILLIAM H. WELCH	1906-34
WALTER A. JESSUP	1938-44	ANDREW D. WHITE	1902-03
SAMUEL P. LANGLEY	1904-06	EDWARD D. WHITE	1902-03
CHARLES A. LINDBERGH	1934-39	HENRY WHITE	1913-27
WILLIAM LINDSAY	1902-09	GEORGE W. WICKERSHAM	1909-36
HENRY CABOT LODGE	1914-24	ROBERT S. WOODWARD	1905-24
SETH LOW	1902-16	CARROLL D. WRIGHT	1902-08
WAYNE MACVEAGH	1902-07		

Besides the names enumerated above, the following were ex-officio members of the Board of Trustees under the original charter, from the date of organization until April 28, 1904: the President of the United States, the President of the Senate, the Speaker of the House of Representatives, the Secretary of the Smithsonian Institution, the President of the National Academy of Sciences.

STAFF OF INVESTIGATORS FOR THE YEAR 1948

ASTRONOMY

MOUNT WILSON OBSERVATORY

813 Santa Barbara Street, Pasadena 4, California

Organized in 1904; George E. Hale, Director 1904-1923, Honorary Director 1923-1936; Walter S. Adams, Director 1924-1945.

IRA S. BOWEN, *Director*
WALTER BAADE
*HAROLD D. BABCOCK
HORACE W. BABCOCK
EDISON HOGE
EDWIN P. HUBBLE
MILTON L. HUMASON
*ALFRED H. JOY
†ROBERT B. KING

PAUL W. MERRILL
RUDOLPH MINKOWSKI
SETH B. NICHOLSON
EDISON PETTIT
ROBERT S. RICHARDSON
ROSCOE F. SANFORD
OLIN C. WILSON
RALPH E. WILSON

MOUNT WILSON AND PALOMAR OBSERVATORIES

Unified operation of the Mount Wilson and Palomar Observatories began on April 1, 1948, the effective date of the appointment by the Presidents of the Carnegie Institution of Washington and California Institute of Technology of Dr. Ira S. Bowen as Director of the Mount Wilson and Palomar Observatories.

IRA S. BOWEN, *Director*
WALTER BAADE
HORACE W. BABCOCK
JESSE L. GREENSTEIN
EDWIN P. HUBBLE
MILTON L. HUMASON
JOSEF J. JOHNSON
ALFRED H. JOY
PAUL W. MERRILL

RUDOLPH L. MINKOWSKI
SETH B. NICHOLSON
EDISON PETTIT
ROBERT S. RICHARDSON
ROSCOE F. SANFORD
OLIN C. WILSON
RALPH E. WILSON
FRITZ ZWICKY

TERRESTRIAL SCIENCES

GEOPHYSICAL LABORATORY

2801 Upton Street, N.W., Washington 8, D. C.

Organized in 1906, opened in 1907; Arthur L. Day, Director 1909-1936

LEASON H. ADAMS, *Director*
NORMAN L. BOWEN
JOHN S. BURLEW
GORDON L. DAVIS
JOSEPH L. ENGLAND
ROY W. GORANSON
JOSEPH W. GREIG
FRANK C. KRACEK
GEORGE W. MOREY

JOHN F. SCHAIRER
O. FRANK TUTTLE
WILLIAM D. URRY
EMANUEL G. ZIES
Visiting Investigators
FELIX CHAYES
MACKENZIE LAWRENCE KEITH
KAARLO J. NEUVONEN
THURF G. SAHAMA

* Retired in 1948.

† Resigned in 1948.

TERRESTRIAL SCIENCES—*Continued*

DEPARTMENT OF TERRESTRIAL MAGNETISM

5241 Broad Branch Road, N.W., Washington 15, D. C.

Organized in 1904; Louis A. Bauer, Director 1904–1929; John A. Fleming, Acting Director 1929–1934, Director 1935–1946.

MERLE A. TUVE, *Director*
PHILIP H. ABELSON
LLOYD V. BERKNER
DEAN B. COWIE
JOHN B. DOAK
SCOTT E. FORBUSH
*OLIVER H. GISH
NORMAN P. HEYDENBURG
†ELLIS A. JOHNSON
‡PAUL G. LEDIG
PAUL L. MOATS
WILFRED C. PARKINSON
RICHARD B. ROBERTS
WILLIAM J. ROONFY
WALTER E. SCOTT

WILLIAM F. STEINER
HOWARD E. TATEL
OSCAR W. TORRESON
ERNEST H. VESTINE
GEORGE R. WAIT
HARRY W. WELLS

Visiting Investigators

HUGH H. DARBY
WILLIAM R. DURYEE
VINCENT C. A. FERRARO
JOHN W. GRAHAM
THOMAS MURPHY
ARTHUR T. NENS
MANUEL S. VALLARTA

BIOLOGICAL SCIENCES

DIVISION OF PLANT BIOLOGY

Central Laboratory, Stanford, California

Desert Laboratory, opened in 1903, became headquarters of Department of Botanical Research in 1905. Name changed to Laboratory for Plant Physiology in 1923; reorganized in 1928 as Division of Plant Biology, including Ecology. Herman A. Spoehr, Chairman 1927–1930 and 1931–1947.

C. STACY FRENCH, *Director*
JENS C. CLAUSEN
WILLIAM M. HIESEY
DAVID D. KECK

HAROLD W. MILNER
JAMES H. C. SMITH
HERMAN A. SPOEHR
HAROLD H. STRAIN

DEPARTMENT OF EMBRYOLOGY

Wolfe and Madison Streets, Baltimore 5, Maryland

Organized in 1914; Franklin P. Mall, Director 1914–1917; George L. Streeter, Director 1918–1940

GEORGE W. CORNER, *Director*
ROBERT K. BURNS
LOUIS B. FLEXNER
CHESTER H. HEUSER, *Curator of the Embryological Collection*

SAMUEL R. M. REYNOLDS
DAVID B. TYLER

DEPARTMENT OF GENETICS

Cold Spring Harbor, Long Island, New York

Station for Experimental Evolution, opened in 1904, combined with Eugenics Record Office in 1921 to form Department of Genetics. Charles B. Davenport, Director 1904–1934; Albert F. Blakeslee, Director 1935–1941.

MILISLAV DEMEREC, *Director*
BERWIND P. KAUFMANN
EDWIN C. MACDOWELL
BARBARA MCCLINTOCK

MARGARET R. McDONALD
Research Associate
ERNEST W. CASPARI

* Retired in 1948.

† On leave of absence.

‡ Resigned in 1948.

HISTORICAL RESEARCH

DIVISION OF HISTORICAL RESEARCH

10 Frisbie Place, Cambridge 38, Massachusetts

Department of Historical Research organized in 1903; Andrew C. McLaughlin, Director 1903-1905; J. Franklin Jameson, Director 1905-1928. In 1930 this Department was incorporated as a section of United States history in a new Division of Historical Research.

ALFRED V. KIDDER, *Chairman*

HARRY E. D. POLLOCK, *Executive Officer*

MARGARET W. HARRISON, *Editor*

*SYLVANUS G. MORLEY

EARL H. MORRIS

ALEXANDER POGO

TATIANA PROSKOURIAKOFF

RALPH L. ROYS

KARL RUPPERT

GEORGE SARTON

ANNA O. SHEPARD

EDWIN M. SHOOK

A. LEDYARD SMITH

ROBERT E. SMITH

GUSTAV STROMSVIK

J. ERIC S. THOMPSON

RESEARCH ASSOCIATES

RESEARCH ASSOCIATES ENGAGED IN POST-RETIREMENT STUDIES

WALTER S. ADAMS, Astronomy

HERBERT E. MERWIN, Geophysics

†GEORGE L. STRUTTER, Embryology

RESEARCH ASSOCIATES CONNECTED WITH OTHER INSTITUTIONS

V. BJERKNES (University of Oslo), Meteorology

JOSEPH C. BOYCE (New York University), Physics

RALPH W. CHANEY (University of California), Paleobotany

TH. DORZHANSKY (Columbia University), Genetics

JOSEPH GILLMAN (Johannesburg Medical School), Embryology

FRANK T. GUCKER, JR. (Indiana University), Chemistry

ROSS G. HARRISON (Yale University), Biology

ARTHUR T. HERTIG (Boston Lying-in Hospital), Embryology

E. A. LOWE (The Institute for Advanced Study), Paleography

ROBERT REDFIELD (University of Chicago), Anthropology

FRANCE V. SCHOLES (University of New Mexico), History

JOEL STEBBINS (University of Wisconsin), Astronomy

SOL TAX (University of Chicago), Ethnology

* Deceased September 2, 1948.

† Deceased July 27, 1948.

OFFICES OF ADMINISTRATION

Office of the President

VANNILVAR BUSH, *President*
PAUL A. SCHERER, *Executive Officer*
SAMUEL CALLAWAY, *President's Secretary*

Office of Publications and Public Relations

FREDERICK G. FASSETT, JR., *Director*
AILENE J. BAUER, *Assistant to the Director*
DOROTHY R. SWIFT, *Editor*

Adviser on International Scientific Relations

JOHN A. FLEMING

Office of the Bursar

EARLE B. BIESECKER, *Bursar*
J. STANLEY LINGEBACH, *Assistant Bursar*
JAMES F. SULLIVAN, *Assistant to the Bursar*

Investment Office (New York City)

PARKER MONROE, *Investment Officer*
RICHARD F. F. NICHOLS, *Assistant Investment Officer*

ORGANIZATION, PLAN, AND SCOPE

The Carnegie Institution of Washington was founded by Andrew Carnegie, January 28, 1902, when he gave to a board of trustees an endowment of registered bonds of the par value of ten million dollars. To this fund an addition of two million dollars was made by Mr. Carnegie on December 10, 1907, and a further addition of ten million dollars was made by him on January 19, 1911. Furthermore, the income of a reserve fund of about three million dollars, accumulated in accordance with the founder's specifications in 1911, is now available for general use, and in recent years a total of ten million dollars has been paid by the Carnegie Corporation of New York as increase to the Endowment Fund of the Institution. The Institution was originally organized under the laws of the District of Columbia and incorporated as the *Carnegie Institution*, articles of incorporation having been executed on January 4, 1902. The Institution was reincorporated, however, by an act of the Congress of the United States, approved April 28, 1904, under the title of the *Carnegie Institution of Washington*. (See existing Articles of Incorporation on following pages.)

Organization under the new Articles of Incorporation was effected May 18, 1904, and the Institution was placed under the control of a board of twenty-four trustees, all of whom had been members of the original corporation. The trustees meet annually in December to consider the affairs of the Institution in general, the progress of work already undertaken, and the initiation of new projects, and to make the necessary appropriations for the ensuing year. During the intervals between the meetings of the trustees the affairs of the Institution are conducted by an Executive Committee chosen by and from the Board of Trustees and acting through the President of the Institution as chief executive officer.

The Articles of Incorporation of the Institution declare in general "that the objects of the corporation shall be to encourage, in the broadest and most liberal manner, investigation, research, and discovery, and the application of knowledge to the improvement of mankind."

The Institution is essentially an operating organization. It attempts to advance fundamental research in fields not normally covered by the activities of other agencies, and to concentrate its attention upon specific problems, with the idea of shifting attack from time to time to meet the more pressing needs of research as they develop with increase of knowledge. Some of these problems require the collaboration of several investigators, special equipment, and continuous effort. Many close relations exist among activities of the Institution, and a type of organization representing investigations in astronomy, in terrestrial sciences, in biological sciences, and in historical research has been effected. Conference groups on various subjects have played a part in bringing new vision and new methods to bear upon many problems. Constant efforts are made to facilitate interpretation and application of results of research activities of the Institution, and an Office of Publications and Public Relations provides means for appropriate publication.

ARTICLES OF INCORPORATION

PUBLIC No. 260. An Act to incorporate the Carnegie Institution of Washington.

Be it enacted by the Senate and House of Representatives of the United States of America in Congress assembled, That the persons following, being persons who are now trustees of the Carnegie Institution, namely, Alexander Agassiz, John S. Billings, John L. Cadwalader, Cleveland H. Dodge, William N. Frew, Lyman J. Gage, Daniel C. Gilman, John Hay, Henry L. Higginson, William Wirt Howe, Charles L. Hutchinson, Samuel P. Langley, William Lindsay, Seth Low, Wayne MacVeagh, Darius O. Mills, S. Weir Mitchell, William W. Morrow, Ethan A. Hitchcock, Elihu Root, John C. Spooner, Andrew D. White, Charles D. Walcott, Carroll D. Wright, their associates and successors, duly chosen, are hereby incorporated and declared to be a body corporate by the name of the Carnegie Institution of Washington and by that name shall be known and have perpetual succession, with the powers, limitations, and restrictions herein contained.

SEC. 2. That the objects of the corporation shall be to encourage, in the broadest and most liberal manner, investigation, research, and discovery, and the application of knowledge to the improvement of mankind; and in particular—

(a) To conduct, endow, and assist investigation in any department of science, literature, or art, and to this end to cooperate with governments, universities, colleges, technical schools, learned societies, and individuals.

(b) To appoint committees of experts to direct special lines of research.

(c) To publish and distribute documents.

(d) To conduct lectures, hold meetings, and acquire and maintain a library.

(e) To purchase property, real or personal, and construct such building or buildings as may be necessary to carry on the work of the corporation.

(f) In general, to do and perform all things necessary to promote the objects of the institution, with full power, however, to the trustees hereinafter appointed and their successors from time to time to modify the conditions and regulations under which the work shall be carried on, so as to secure the application of the funds in the manner best adapted to the conditions of the time, provided that the objects of the corporation shall at all times be among the foregoing or kindred thereto.

SEC. 3. That the direction and management of the affairs of the corporation and the control and disposal of its property and funds shall be vested in a board of trustees, twenty-two in number, to be composed of the following individuals: Alexander Agassiz, John S. Billings, John L. Cadwalader, Cleveland H. Dodge, William N. Frew, Lyman J. Gage, Daniel C. Gilman, John Hay, Henry L. Higginson, William Wirt Howe, Charles L. Hutchinson, *Samuel P. Langley*, William Lindsay, Seth Low, Wayne MacVeagh, Darius O. Mills, S. Weir Mitchell, William W. Morrow, *Ethan A. Hitchcock*, Elihu Root, John C. Spooner, Andrew D. White, Charles D. Walcott, Carroll D. Wright, who shall constitute the first board of trustees. The board of trustees shall have power from time to time to increase its membership to not more than twenty-seven members. Vacancies occasioned by death, resignation, or otherwise shall be filled by the remaining trustees in such manner as the by-laws

ARTICLES OF INCORPORATION

shall prescribe; and the persons so elected shall thereupon become trustees and also members of the said corporation. The principal place of business of the said corporation shall be the city of Washington, in the District of Columbia.

SEC. 4. That such board of trustees shall be entitled to take, hold, and administer the securities, funds, and property so transferred by said Andrew Carnegie to the trustees of the Carnegie Institution and such other funds or property as may at any time be given, devised, or bequeathed to them, or to such corporation, for the purposes of the trust; and with full power from time to time to adopt a common seal, to appoint such officers, members of the board of trustees or otherwise, and such employees as may be deemed necessary in carrying on the business of the corporation, at such salaries or with such remuneration as they may deem proper; and with full power to adopt by-laws from time to time and such rules or regulations as may be necessary to secure the safe and convenient transaction of the business of the corporation; and with full power and discretion to deal with and expend the income of the corporation in such manner as in their judgment will best promote the objects herein set forth and in general to have and use all powers and authority necessary to promote such objects and carry out the purposes of the donor. The said trustees shall have further power from time to time to hold as investments the securities hereinabove referred to so transferred by Andrew Carnegie, and any property which has been or may be transferred to them or such corporation by Andrew Carnegie or by any other person, persons, or corporation, and to invest any sums or amounts from time to time in such securities and in such form and manner as are permitted to trustees or to charitable or literary corporations for investment, according to the laws of the States of New York, Pennsylvania, or Massachusetts, or in such securities as are authorized for investment by the said deed of trust so executed by Andrew Carnegie, or by any deed of gift or last will and testament to be hereafter made or executed.

SEC. 5. That the said corporation may take and hold any additional donations, grants, devises, or bequests which may be made in further support of the purposes of the said corporation, and may include in the expenses thereof the personal expenses which the trustees may incur in attending meetings or otherwise in carrying out the business of the trust, but the services of the trustees as such shall be gratuitous.

SEC. 6. That as soon as may be possible after the passage of this Act a meeting of the trustees hereinbefore named shall be called by Daniel C. Gilman, John S. Billings, Charles D. Walcott, S. Weir Mitchell, John Hay, Elihu Root, and Carroll D. Wright, or any four of them, at the city of Washington, in the District of Columbia, by notice served in person or by mail addressed to each trustee at his place of residence; and the said trustees, or a majority thereof, being assembled, shall organize and proceed to adopt by-laws, to elect officers and appoint committees, and generally to organize the said corporation; and said trustees herein named, on behalf of the corporation hereby incorporated, shall thereupon receive, take over, and enter into possession, custody, and management of all property, real or personal, of the corporation heretofore known as the Carnegie Institution, incorporated, as hereinbefore set forth under "An Act to establish a Code of Law for the District of Columbia, January fourth, nineteen hundred and two," and to all its rights, contracts, claims,

CARNEGIE INSTITUTION OF WASHINGTON

and property of any kind or nature; and the several officers of such corporation, or any other person having charge of any of the securities, funds, real or personal, books, or property thereof, shall, on demand, deliver the same to the said trustees appointed by this Act or to the persons appointed by them to receive the same; and the trustees of the existing corporation and the trustees herein named shall and may take such other steps as shall be necessary to carry out the purposes of this Act.

SEC. 7. That the rights of the creditors of the said existing corporation known as the Carnegie Institution shall not in any manner be impaired by the passage of this Act, or the transfer of the property hereinbefore mentioned, nor shall any liability or obligation for the payment of any sums due or to become due, or any claim or demand, in any manner or for any cause existing against the said existing corporation, be released or impaired; but such corporation hereby incorporated is declared to succeed to the obligations and liabilities and to be held liable to pay and discharge all of the debts, liabilities, and contracts of the said corporation so existing to the same effect as if such new corporation had itself incurred the obligation or liability to pay such debt or damages, and no such action or proceeding before any court or tribunal shall be deemed to have abated or been discontinued by reason of the passage of this Act.

SEC. 8. That Congress may from time to time alter, repeal, or modify this Act of incorporation, but no contract or individual right made or acquired shall thereby be divested or impaired.

SEC. 9. That this Act shall take effect immediately.

Approved, April 28, 1904

BY-LAWS OF THE INSTITUTION

Adopted December 13, 1904. Amended December 13, 1910, December 13, 1912, December 10, 1937, December 15, 1939, December 13, 1940, December 18, 1942, and December 12, 1947

ARTICLE I

THE TRUSTEES

1. The Board of Trustees shall consist of twenty-four members, with power to increase its membership to not more than twenty-seven members. The Trustees shall hold office continuously and not for a stated term.
2. In case any Trustee shall fail to attend three successive annual meetings of the Board he shall thereupon cease to be a Trustee.
3. No Trustee shall receive any compensation for his services as such.
4. All vacancies in the Board of Trustees shall be filled by the Trustees by ballot at an annual meeting, but no person shall be declared elected unless he receives the votes of two-thirds of the Trustees present.

ARTICLE II

OFFICERS OF THE BOARD

1. The officers of the Board shall be a Chairman of the Board, a Vice-Chairman, and a Secretary, who shall be elected by the Trustees, from the members of the Board, by ballot to serve for a term of three years. All vacancies shall be filled by the Board for the unexpired term; provided, however, that the Executive Committee shall have power to fill a vacancy in the office of Secretary to serve until the next meeting of the Board of Trustees.
2. The Chairman shall preside at all meetings and shall have the usual powers of a presiding officer.
3. The Vice-Chairman, in the absence or disability of the Chairman, shall perform the duties of the Chairman.
4. The Secretary shall issue notices of meetings of the Board, record its transactions, and conduct that part of the correspondence relating to the Board and to his duties.

ARTICLE III

EXECUTIVE ADMINISTRATION

The President

1. There shall be a President who shall be elected by ballot by, and hold office during the pleasure of, the Board, who shall be the chief executive officer of the Institution. The President, subject to the control of the Board and the Executive Committee, shall have general charge of all matters of administration and supervision of all arrangements for research and other work undertaken by the Institution or with its funds. He shall prepare and submit to the Board of Trustees and to the Executive

CARNEGIE INSTITUTION OF WASHINGTON

Committee plans and suggestions for the work of the Institution, shall conduct its general correspondence and the correspondence with applicants for grants and with the special advisers of the Committee, and shall present his recommendations in each case to the Executive Committee for decision. All proposals and requests for grants shall be referred to the President for consideration and report. He shall have power to remove, appoint, and, within the scope of funds made available by the Trustees, provide for compensation of subordinate employees and to fix the compensation of such employees within the limits of a maximum rate of compensation to be established from time to time by the Executive Committee. He shall be *ex officio* a member of the Executive Committee.

2. He shall be the legal custodian of the seal and of all property of the Institution whose custody is not otherwise provided for. He shall sign and execute on behalf of the corporation all contracts and instruments necessary in authorized administrative and research matters and affix the corporate seal thereto when necessary, and may delegate the performance of such acts and other administrative duties in his absence to the Executive Officer. He may execute all other contracts, deeds, and instruments on behalf of the corporation and affix the seal thereto when expressly authorized by the Board of Trustees or Executive Committee. He may, within the limits of his own authorization, delegate to the Executive Officer authority to act as custodian of and affix the corporate seal. He shall be responsible for the expenditure and disbursement of all funds of the Institution in accordance with the directions of the Board and of the Executive Committee, and shall keep accurate accounts of all receipts and disbursements. Following approval by the Executive Committee he shall transmit to the Board of Trustees before its annual meeting in December a written report of the operations and business of the Institution for the preceding fiscal year with his recommendations for work and appropriations for the succeeding calendar year.

3. He shall attend all meetings of the Board of Trustees.

4. There shall be an officer designated Executive Officer who shall be appointed by and hold office at the pleasure of the President, subject to the approval of the Executive Committee. His duties shall be to assist and act for the President as the latter may duly authorize and direct.

5. The President shall retire from office at the end of the calendar year in which he becomes sixty-five years of age.

ARTICLE IV

MEETINGS

1. The annual meeting of the Board of Trustees shall be held in the city of Washington, in the District of Columbia, on the first Friday following the second Thursday of December in each year unless the date and place of meeting are otherwise ordered by the Executive Committee.

2. Special meetings of the Board may be called by the Executive Committee by notice served personally upon, or mailed to the usual address of, each Trustee twenty days prior to the meeting.

3. Special meetings shall, moreover, be called in the same manner by the Chairman upon the written request of seven members of the Board.

BY-LAWS OF THE INSTITUTION

ARTICLE V

COMMITTEES

1. There shall be the following standing Committees, *viz.* an Executive Committee, a Finance Committee, an Auditing Committee, and a Nominating Committee.
2. All vacancies occurring in the Executive Committee, the Finance Committee, the Auditing Committee, and the Nominating Committee shall be filled by the Trustees at the next regular meeting. In case of vacancy in the Finance Committee, the Auditing Committee, or the Nominating Committee, upon request of the remaining members of such committee, the Executive Committee may fill such vacancy by appointment until the next meeting of the Board of Trustees.
3. The terms of all officers and of all members of committees, as provided for herein, shall continue until their successors are elected or appointed.

Executive Committee

4. The Executive Committee shall consist of the Chairman and Secretary of the Board of Trustees and the President of the Institution *ex officio* and, in addition, five trustees to be elected by the Board by ballot for a term of three years, who shall be eligible for re-election. Any member elected to fill a vacancy shall serve for the remainder of his predecessor's term.
5. The Executive Committee shall, when the Board is not in session and has not given specific directions, have general control of the administration of the affairs of the corporation and general supervision of all arrangements for administration, research, and other matters undertaken or promoted by the Institution. It shall also submit to the Board of Trustees a printed or typewritten report of each of its meetings, and at the annual meeting shall submit to the Board a report for publication.
6. The Executive Committee shall have power to authorize the purchase, sale, exchange, or transfer of real estate.

Finance Committee

7. The Finance Committee shall consist of five members to be elected by the Board of Trustees by ballot for a term of three years.
8. The Finance Committee shall have custody of the securities of the corporation and general charge of its investments and invested funds, and shall care for and dispose of the same subject to the directions of the Board of Trustees. It shall have power to authorize the purchase, sale, exchange, or transfer of securities and to delegate this power. It shall consider and recommend to the Board from time to time such measures as in its opinion will promote the financial interests of the Institution, and shall make a report at each meeting of the Board.

Auditing Committee

9. The Auditing Committee shall consist of three members to be elected by the Board of Trustees by ballot for a term of three years.

CARNEGIE INSTITUTION OF WASHINGTON

10. Before each annual meeting of the Board of Trustees, the Auditing Committee shall cause the accounts of the Institution for the preceding fiscal year to be audited by public accountants. The accountants shall report to the Committee, and the Committee shall present said report at the ensuing annual meeting of the Board with such recommendations as the Committee may deem appropriate.

Nominating Committee

11. The Nominating Committee shall consist of the Chairman of the Board of Trustees *ex officio* and, in addition, three trustees to be elected by the Board by ballot for a term of three years, who shall not be eligible for re-election until after the lapse of one year. Any member elected to fill a vacancy shall serve for the remainder of his predecessor's term, provided that of the Nominating Committee first elected after adoption of this By-Law one member shall serve for one year, one member shall serve for two years, and one member shall serve for three years, the Committee to determine the respective terms by lot.

12. Sixty days prior to an annual meeting of the Board the Nominating Committee shall notify the Trustees by mail of the vacancies to be filled in membership of the Board. Each Trustee may submit nominations for such vacancies. Nominations so submitted shall be considered by the Nominating Committee, and ten days prior to the annual meeting the Nominating Committee shall submit to members of the Board by mail a list of the persons so nominated, with its recommendations for filling existing vacancies on the Board and its Standing Committees. No other nominations shall be received by the Board at the annual meeting except with the unanimous consent of the Trustees present.

ARTICLE VI

FINANCIAL ADMINISTRATION

1. No expenditure shall be authorized or made except in pursuance of a previous appropriation by the Board of Trustees, or as provided in Article V, paragraph 8, hereof.

2. The fiscal year of the Institution shall commence on the first day of July in each year.

3. The Executive Committee shall submit to the annual meeting of the Board a full statement of the finances and work of the Institution for the preceding fiscal year and a detailed estimate of the expenditures of the succeeding calendar year.

4. The Board of Trustees, at the annual meeting in each year, shall make general appropriations for the ensuing calendar year; but nothing contained herein shall prevent the Board of Trustees from making special appropriations at any meeting.

5. The Executive Committee shall have general charge and control of all appropriations made by the Board. Following the annual meeting each year, the Executive Committee may make allotment of funds for the period from January 1 to termination of the fiscal year on June 30. It may also make allotment of funds for the period from July 1 to December 31 in advance of July 1. The Committee shall, however, have full authority for allotment of available funds to meet necessary

BY-LAWS OF THE INSTITUTION

expenditures by other methods, if desirable, and for transfer of balances to meet special needs. It shall make provision for outstanding obligations and for reversion of unexpended balances at termination of the fiscal year.

6. The securities of the Institution and evidences of property, and funds invested and to be invested, shall be deposited in such safe depository or in the custody of such trust company and under such safeguards as the Finance Committee shall designate, subject to directions of the Board of Trustees. Income of the Institution available for expenditure shall be deposited in such banks or depositories as may from time to time be designated by the Executive Committee.

7. Any trust company entrusted with the custody of securities by the Finance Committee may, by resolution of the Board of Trustees, be made Fiscal Agent of the Institution, upon an agreed compensation, for the transaction of the business coming within the authority of the Finance Committee.

ARTICLE VII

AMENDMENT OF BY-LAWS

1. These by-laws may be amended at any annual or special meeting of the Board of Trustees by a two-thirds vote of the members present, provided written notice of the proposed amendment shall have been served personally upon, or mailed to the usual address of, each member of the Board twenty days prior to the meeting.

ABSTRACT OF MINUTES OF THE FIFTIETH MEETING OF THE BOARD OF TRUSTEES

The annual meeting of the Board of Trustees was held in Washington, D. C., in the Board Room of the Administration Building, on Friday, December 10, 1948. It was called to order at 10:40 A.M. by the Chairman, Mr. Gifford.

Upon roll call, the following Trustees responded: James F. Bell, Robert Woods Bliss, Lindsay Bradford, W. Cameron Forbes, Walter S. Gifford, Ernest O. Lawrence, Alfred L. Loomis, Roswell Miller, Henry S. Morgan, Seeley G. Mudd, Henning W. Prentis, Jr., Elihu Root, Jr., Henry R. Shepley, Charles P. Taft, James W. Wadsworth, and Lewis H. Weed. The President of the Institution, Vannevar Bush, was also present.

The minutes of the forty-ninth meeting were approved as printed and submitted to the members of the Board.

Reports of the President, the Executive Committee, the Finance Committee, the Auditor, the Auditing Committee, and the Nominating Committee, and of the heads of Divisions and Departments and Research Associates of the Institution were presented and considered.

The sum of \$1,621,978.67 was appropriated for expenditure by the Institution under the general charge and control of the Executive Committee.

Vacancies in the membership of the Board of Trustees caused by the deaths of Gordon S. Rentschler and Richard P. Strong and by the resignation of Frederic C. Walcott were filled by the election of James Forrestal, Robert A. Lovett, and William I. Myers.

The following officers of the Board were re-elected for a period of three years: Walter S. Gifford, Chairman, Elihu Root, Jr., Vice-Chairman, and Lewis H. Weed, Secretary. Robert Woods Bliss, Elihu Root, Jr., and Henry R. Shepley were re-elected members of the Executive Committee for a period of three years, and Frank B. Jewett and Henry S. Morgan were elected as members of this committee for the terms ending in 1950 to succeed Mr. Rentschler and Mr. Walcott. Alfred L. Loomis, Henry S. Morgan, and Elihu Root, Jr. were re-elected members of the Finance Committee for a period of three years. Frederic A. Delano, Chairman, Homer L. Ferguson, and James W. Wadsworth were re-elected members of the Auditing Committee for a period of three years. James F. Bell was elected a member of the Nominating Committee for a period of three years, succeeding Frank B. Jewett.

A Manual of Fiscal Procedures was approved.

The meeting adjourned at 12:15 P.M.

REPORT OF THE EXECUTIVE COMMITTEE

FOR THE PERIOD ENDED JUNE 30, 1948

To the Trustees of the Carnegie Institution of Washington:

GENTLEMEN: Article V, Section 5 of the By-Laws provides that the Executive Committee shall submit, at the annual meeting of the Board of Trustees, a report for publication; and Article VI, Section 3, provides that the Executive Committee shall also submit, at the same time, a full statement of the finances and work of the Institution for the preceding fiscal year and a detailed estimate of the expenditures of the succeeding calendar year. The Executive Committee, therefore, respectfully submits this report for the fiscal period ended June 30, 1948.

During the eight-month fiscal period between October 31, 1947 and June 30, 1948, the Executive Committee held three meetings, printed reports of which have been mailed to each Trustee and constitute a part of this report.

A statement of activities of the Institution is contained in the report of the President, which has been considered and approved by the Executive Committee.

The detailed estimate of expenditures for the calendar year 1949 contained in the report of the President has been considered by the Executive Committee, which has approved the recommendations of the President in respect thereto and has provisionally approved the budget estimates based thereon.

The Board of Trustees at its meeting of December 12, 1947 appointed the firm of Haskins & Sells to audit the accounts of the Institution for the fiscal year ending June 30, 1948. The report of the Auditor, including a balance sheet showing assets and liabilities of the Institution on June 30, 1948, is submitted as a part of the report of the Executive Committee.

In addition to the report of the Auditor there is also submitted a financial statement for the fiscal year ended June 30, 1948, showing funds available for expenditure and amounts allotted by the Executive Committee, and the customary statement of aggregate receipts and disbursements, together with a schedule of real estate and equipment. These statements together with the tables in the Auditor's report comprise the statement of the finances of the Institution.

Three vacancies exist in the membership of the Board of Trustees resulting from the death in March 1948 of Gordon S. Rentschler, the death in July 1948 of Richard P. Strong, and the resignation of Frederic C. Walcott in May 1948.

Terms of the following officers of the Board of Trustees will end at the annual meeting in December: Mr. Gifford, Chairman of the Board; Mr. Root, Vice-Chairman of the Board; and Mr. Weed, Secretary of the Board. Terms of Messrs. Bliss, Root, and Shepley as members of the Executive Committee (and two vacancies have resulted from the death of Mr. Rentschler and the resignation of Mr. Walcott); of Messrs. Loomis, Morgan, and Root as members of the Finance Committee; and of

Messrs. Delano, Ferguson, and Wadsworth as members of the Auditing Committee will end at the annual meeting. The term of Mr. Jewett, Chairman, as a member of the Nominating Committee will also end at the annual meeting.

WALTER S. GIFFORD, *Chairman*

ROBERT WOODS BLISS

VANNEVAR BUSH

ELIHU ROOT, JR.

HENRY R. SHEPLEY

LEWIS H. WEED

October 21, 1948

FINANCIAL STATEMENT FOR FISCAL YEAR ENDING JUNE 30, 1948*

Departmental Research Operations:

	Balances unallotted Oct. 31, 1947	Trustees' appropriations Jan.-June 1948	Transfers and other credits	Total available	Executive Committee allotments	Transfers by Executive Committee	Unallotted balances June 30, 1948
Embryology.....	\$48,420.00	\$695.00	\$49,115.00	\$49,115.00
Genetics.....	61,655.00	61,655.00	61,655.00
Geophysical Laboratory.....	96,150.00	2,000.00	98,150.00	98,150.00
Historical Research.....	62,833.50	1,330.00	64,163.50	64,163.50
Mount Wilson Observatory.....	120,075.00	1,500.00	121,575.00	121,575.00
Plant Biology.....	39,800.00	2,225.00	42,025.00	42,025.00
Terrestrial Magnetism.....	130,250.00	13,000.00	143,250.00	143,250.00
Research Projects of Limited Tenure.....	\$990.55	990.55	\$990.55
Publication.....	12,705.93	17,200.00	21,353.13	51,259.06	29,680.35	21,578.71
Administration.....	54,000.00	6,375.00	60,375.00	60,375.00
Pension Fund.....	47,500.00	47,500.00	47,500.00
General Contingent Fund.....	98,908.96	41,200.00	307,633.14	447,742.10	81,488.42	\$30,825.00	335,428.68
Carnegie Corporation Emergency Fund...	115,900.41	115,900.41	115,900.41
General Operations.....	31,000.00	31,000.00	31,000.00
	<u>\$228,505.85</u>	<u>\$750,083.50</u>	<u>\$356,111.27</u>	<u>\$1,334,700.62</u>	<u>\$829,977.27</u>	<u>\$30,825.00</u>	<u>\$473,898.35</u>

*End of fiscal year changed from October 31 to June 30.

AGGREGATE CASH RECEIPTS AND DISBURSEMENTS FROM ORGANIZATION, JANUARY 28, 1902, TO JUNE 30, 1948

RECEIPTS

<i>Securities Sold or Redeemed</i>	\$134,631,200.02
<i>Income from Securities and Bank Balances</i>	57,555,826.40
<i>Sales of Publications</i>	392,498.67
<i>Colburn Estate (Bequest)</i>	52,015.74
<i>Harkavy Fund (Gift)</i>	3,050.00
<i>Harriman Fund (Sale of Land)</i>	4,043.70
<i>Teetle Estate (Bequest)</i>	10,839.76
<i>Van Gelder Fund (Bequest)</i>	1,278.58
<i>Carnegie Corporation of New York (Endowment Increase and for Specific Purposes)</i>	13,717,381.24
<i>From Other Organizations and Individuals for Specific Purposes</i>	512,698.98
<i>Pension Fund (Refunds)</i>	100,177.50
<i>General Reserve Fund</i>	74,609.44
<i>Insurance Fund (Refunds)</i>	13,076.02
<i>National Defense Revolving Fund (Refunds and Advances)</i>	3,095,347.20
<i>Administration Building Addition Account, Rentals and Refunds</i>	18,021.09
<i>Employees' Salary Deductions for the Purchase of U. S. Bonds</i>	94,770.90
<i>Miscellaneous Refunds and Receipts</i>	1,196,803.56
	<hr/>
	\$211,473,638.80
	<hr/>

DISBURSEMENTS

<i>Securities Purchased</i>	\$150,442,246.67
<i>Accrued Interest on Securities Purchased</i>	794,651.08
<i>Pension Fund</i>	1,937,063.40
<i>General Reserve Fund</i>	30,477.43
<i>Insurance Fund</i>	140,532.24
<i>Harriman Fund</i>	346.44
<i>Harriet H. Mayor Relief Fund</i>	250.00
<i>Harkavy Fund</i>	221.20
<i>Special Emergency Reserve Fund</i>	63,819.41
<i>National Defense Revolving Fund</i>	3,062,974.97
<i>General Contingent Fund</i>	342,289.26
<i>Carnegie Corporation of New York Emergency Fund</i>	101,444.77
<i>Administration Building and Addition: Construction and Site (Old Building)</i>	309,915.69
<i>Construction (Addition to Administration Bldg.)</i>	416,206.07
<i>Site (Addition to Administration Building)</i>	68,570.96
<i>Miscellaneous Expenditures*</i>	40,825.37
<i>Departmental Research Operations:</i>	
<i>Departments of Research, Buildings and Equipment</i>	3,789,090.25
<i>Departmental Operations</i>	36,981,308.79
<i>Research Projects, Fellowships, Grants, etc.</i>	5,631,901.37
<i>Publication</i>	3,132,923.39
<i>Administration</i>	3,324,415.23
<i>Employees' U. S. Bond Purchases</i>	94,743.15
<i>National Research Council</i>	150,000.00
<i>Miscellaneous</i>	19,904.16
	<hr/>
<i>June 30, 1948, Cash in Banks</i>	\$210,876,121.30
	597,517.50
	<hr/>
	\$211,473,638.80
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*Includes Equipment \$7,206.41, Repairs and Alterations to Old Building \$18,599.29.

REAL ESTATE AND EQUIPMENT, ORIGINAL COST
JUNE 30, 1948

Administration

1530 P Street, N.W., Washington 5, D. C.

Building, site, and equipment.....		\$850,935.91
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Division of Plant Biology

Stanford, California

Buildings and grounds.....	\$74,556 67	
Laboratory.....	45,637.65	
Library.....	22,874 44	
Operating equipment.....	17,516 69	160,585.45

Department of Embryology

Wolfe and Madison Streets, Baltimore 5, Maryland

Library.....	\$7,744 19	
Laboratory.....	33,030 95	
Administration.....	8,810.26	49,585.40

Department of Genetics

Cold Spring Harbor, Long Island, New York

Buildings, grounds, and field	\$302,317.59	
Operating equipment.....	35,020.47	
Laboratory apparatus.....	55,641 99	
Library.....	63,878 03	456,858.08

Geophysical Laboratory

2801 Upton Street, N.W., Washington 8, D. C.

Buildings, library, and operating appliances.....	\$297,337.45	
Laboratory apparatus.....	187,161.56	
Shop equipment.....	21,346 19	505,845.20

Division of Historical Research

10 Frisbie Place, Cambridge 38, Massachusetts

Operating equipment.....	\$28,850.79	
Library.....	11,563.74	40,414 53

Mount Wilson Observatory

813 Santa Barbara Street, Pasadena 4, California

Buildings and grounds.....	\$260,549 56	
Shop equipment.....	53,775.33	
Instruments.....	688,034.96	
Furniture and operating appliances	165,846.94	
Hooker 100-inch reflector.....	644,803 88	1,813,010 67

Department of Terrestrial Magnetism

5241 Broad Branch Road, N.W., Washington 15, D. C.

Buildings, site, and office.....	\$275,386.10	
Survey equipment.....	20,376.18	
Instruments, laboratory, and shop equipment.....	467,028.64	762,790.92
		\$4,640,026.16

HASKINS & SELLS
CERTIFIED PUBLIC ACCOUNTANTS

500 EQUITABLE BUILDING
BALTIMORE 2
MD.

ACCOUNTANTS' CERTIFICATE

*To the Board of Trustees of
Carnegie Institution of Washington*

We have examined the balance sheet (and supporting schedule of investments owned) of Carnegie Institution of Washington as of June 30, 1948 and the related statement of income and expenditures for the eight months ended that date. Our examination was made in accordance with generally accepted auditing standards and included such tests of the accounting records and such other auditing procedures as we considered necessary in the circumstances, except that we did not examine the records in support of expenditures made (approximately \$250,000) by the branch offices of the Institution, but reviewed internal audit reports of the Bursar covering examinations of the branch records during the period.

In our opinion, subject to the exception stated above with respect to the limitation of the scope of our examination, the accompanying balance sheet and statement of income and expenditures present fairly the financial condition of the Institution at June 30, 1948 and the results of its operations for the eight months ended that date.

HASKINS & SELLS

November 5, 1948

BALANCE SHEET JUNE 30, 1948

ASSETS		LIABILITIES	
<i>Current Funds:</i>		<i>Current Funds:</i>	
General:		General:	
Cash in banks and on hand	\$348,152.75	Employees' U. S. bond deductions	\$27.75
Advances—Departmental Research Operations	14,448.32	Reserved for unexpended appropriations:	
Accounts receivable—U. S. Government	430.00	Departmental Research Operations	\$129,113.07
Accounts receivable—other	745.13	Research Projects, Fellowships, Grants, etc.	166,892.08
Inventory books	157,102.53	Publication	53,190.56
Deferred charges	18,136.40	Administration	6,179.85
Insurance premiums			355,325.56
Dues, Endowment and Other Special Funds	463,127.74		
	\$1,002,142.87	Reserved for valuation of books and accounts receivable	157,766.16
		Reserved for General Contingent Fund	351,460.18
		Reserved for Carnegie Corporation Emergency Fund	115,900.41
		Reserved for special reconversion expenses	12,630.81
		Reserved for general operations	9,032.00
			\$1,002,142.87
Restricted:		Restricted:	
Cash in banks	85,206.75	Harriman Fund—income account	85,128.07
	\$1,087,349.62	Harkavy Fund—income account	78.68
			\$1,087,349.62
<i>Endowment and Other Special Funds:</i>		<i>Endowment and Other Special Funds:</i>	
Cash in banks	\$191,355.60	Due to Current Funds	\$463,127.74
Securities (approximate market value \$44,179,936):		Capital Funds:	
U. S. Government bonds	\$9,530,469.40	Endowment Fund	\$32,000,000.00
Foreign and International Bank bonds	854,008.76	Capital Reserve Fund	5,617,417.32
Public utility bonds	2,106,035.38	Colburn Fund	103,310.80
Communication bonds	1,176,403.96	Harriman Fund	304,043.70
Railroad bonds	274,484.45	Teepie Fund	10,839.76
Railroad equipment trusts	877,386.21	Harkavy Fund	2,828.80
Industrial and miscellaneous bonds	5,240,508.45	Van Gelder Fund	1,278.58
Preferred stocks	4,640,788.86	Special Funds:	
Common stocks	15,857,291.09	Pension Fund	187,974.09
	40,557,376.56	Harriet H. Mayor Relief Fund	9,750.00
	40,748,732.16	General Reserve Fund	2,048,161.37
			40,285,604.42
			40,748,732.16
<i>Plant Funds:</i>		<i>Plant Funds:</i>	
Invested in real estate and equipment:		Income invested in plant*	\$4,465,538.09
Office of Administration	\$850,935.91	Harriman property (gift)	134,418.07
Departments of Research	3,789,090.25	Harkavy property (gift)	2,070.00
	4,640,026.16	Solar Laboratory (Hale gift equipment valued at \$22,000, included above*)	38,000.00
	\$46,476,107.94		\$46,476,107.94

STATEMENT OF OPERATING INCOME AND EXPENDITURES, NOVEMBER 1, 1947 TO JUNE 30, 1948

INCOME (8 months):*

Interest and dividends on securities.....	\$1,062,714.38	
Less—Amortization of bond premiums.....	8,519.29	
		<u>\$1,054,195.09</u>

Sales of publications.....	\$6,267.23	
American Cancer Society—grants.....	8,931.04	
Carnegie Corporation of New York—grant.....	12,000.00	
Dormitory and mess hall.....	7,942.66	
Hall Laboratories, Inc.—grant.....	5,000.00	
Sale of cloud chamber.....	5,703.41	
United Fruit Company—Bonampak project.....	2,500.00	
U. S. Public Health Service—grants.....	9,021.26	
A. F. Zahm—gift.....	2,000.00	
Other credits.....	3,442.00	
		<u>62,807.60</u>

Transfer from General Reserve Fund.....		<u>65,770.04</u>
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TOTAL.....		<u>\$1,182,772.73</u>
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EXPENDITURES (8 months):*

Pension Fund—annuity and insurance.....	\$77,483.35	
Harriman Fund—commissions.....	47.95	

Departmental Research Operations:		
Salaries.....	\$591,713.73	
Operating expenses.....	248,537.61	
Dormitory and mess—salaries.....	2,409.40	
Dormitory and mess—operating expenses...	5,669.25	
		<u>848,329.99</u>

Research Projects, Fellowships, Grants, etc.:		
Salaries.....	\$4,766.88	
Grants and miscellaneous.....	22,478.33	
Fellowship program.....	25,273.75	
		<u>52,518.96</u>

Office of Publications:		
Printing and publishing expenses.....	\$44,811.67	
Office expenses:		
Salaries.....	20,017.83	
Stationery, postage, etc.....	4,066.60	
		<u>68,896.10</u>

Administration.....		<u>78,325.35</u>
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TOTAL.....	\$1,125,601.70	
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Less—Salaries and operating expenses charged to previous appropriations.....	270,803.41	854,798.29
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Excess of income over expenditures.....		\$327,974.44
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Less—Credits to General Reserve Fund and other accounts.....		<u>62,807.60</u>
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Unexpended current income Nov. 1, 1947—June 30, 1948.....		\$265,166.84
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Unexpended current income Oct. 31, 1947.....		<u>139,848.03</u>
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TOTAL.....		<u>\$405,014.87</u>
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DISTRIBUTION OF UNEXPENDED CURRENT INCOME

General Contingent Fund.....	\$305,374.14
General Operations.....	5,732.00
Departments of Research.....	41,372.13
Administration.....	4,201.13
Research Projects, Fellowships, Grants, and Miscellaneous Accounts...	14,041.67
Publications.....	10,000.00
Office of Publications.....	759.67
Pension Fund.....	10,011.17
Harkavy Fund.....	78.68
Harriman Fund.....	13,444.28

TOTAL.....	<u>\$405,014.87</u>
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*End of fiscal year changed from October 31 to June 30.

SCHEDULE OF SECURITIES OWNED JUNE 30, 1948

Aggregate par or nominal value	Description	Maturity	Cost, amortized cost, or value at date acquired	Approximate market value	Net income (8 months)
UNITED STATES GOVERNMENT BONDS					
\$1,100,000	U. S. of America Treasury Bills	7-15-1948	\$1,099,303.34	\$1,099,538
304,000	U. S. of America Treasury 2s	1951-49	304,000.00	306,850	\$3,040.00
312,000	U. S. of America Treasury 2s	1951-49	312,000.00	315,315	6,240.00
200,000	U. S. of America Treasury 2s	1952-50	200,000.00	202,250	2,000.00
4,500,000	U. S. of America Treasury 2s	1954-52	4,500,000.00	4,573,125	90,000.00
800,000	U. S. of America Treasury 2½s	1955-52	800,000.00	819,500	18,000.00
400,000	U. S. of America Treasury 2½s	1959-56	423,434.30*	408,500	4,500.00
1,239,000	U. S. of America Treasury 2½s	1954-52	1,241,731.56*	1,279,635	15,487.50
50,000	U. S. of America Savings Defense "G" 2½s	1953	50,000.00	48,050	1,250.00
50,000	U. S. of America Savings Defense "G" 2½s	1954	50,000.00	47,900	625.00
50,000	U. S. of America Savings Defense "G" 2½s	1954	50,000.00	47,750	625.00
100,000	U. S. of America Savings Defense "G" 2½s	1955	100,000.00	95,200	1,250.00
100,000	U. S. of America Savings Defense "G" 2½s	1956	100,000.00	94,700	1,250.00
100,000	U. S. of America Savings Defense "G" 2½s	1957	100,000.00	94,800	1,250.00
100,000	U. S. of America Savings Defense "G" 2½s	1958	100,000.00	95,600	1,250.00
100,000	U. S. of America Savings Defense "G" 2½s	1959	100,000.00	96,900	1,250.00
Income from bonds sold or redeemed			130,573.22
<u>\$9,505,000</u>	Total U. S. Government	<u>\$9,530,469.40</u>	<u>\$9,625,633</u>	<u>\$278,590.72</u>
FOREIGN AND INTERNATIONAL BANK BONDS					
\$100,000	Australia, Commonwealth of, S. F. 3½s	1956	\$100,000.00	\$90,000	\$1,625.00
50,000	Australia, Commonwealth of, S. F. 3½s	1957	48,750.00	44,500	1,625.00
90,000	Canadian National Ry. Co., 4½s Guar.	1951	90,128.06*	96,300	2,025.00
100,000	Canadian National Ry. Co., 4½s Guar.	1957	112,000.00	113,000	2,250.00
57,000	Canadian National Ry. Co., 5s Guar.	1969	61,213.86*	61,560	1,425.00
35,000	Canadian National Ry. Co., 5s Guar.	1970	37,472.25*	37,800	875.00
100,000	International Bank for Reconstruction and Development, 2½s	1957	100,000.00	97,250	1,125.00
200,000	Shawinigan Water & Power Co., 1st Mtg. & Coll. Tr. S. F. 3s	1971	208,280.00*	198,000	3,000.00
100,000	City of Toronto Cons. Loan Deb. 5s	1949	96,164.59	104,000	5,000.00
<u>\$832,000</u>	Total Foreign and International Bank	<u>\$854,008.76</u>	<u>\$842,410</u>	<u>\$18,950.00</u>

*After deduction for amortization of premiums on bonds purchased subsequent to January 1, 1940.

SCHEDULE OF SECURITIES OWNED JUNE 30, 1948—Continued

Aggregate par or nominal value	Description	Maturity	Cost, amortized cost, or value at date acquired	Approximate market value	Net income (\$ months)
PUBLIC UTILITY BONDS					
\$243,000	Columbus & Southern Ohio Electric Co., 1st Mtg. 3 1/8s.	1970	\$257,461.55*	\$260,010	\$3,948.75
300,000	Consolidated Natural Gas Co., Deb. 2 1/8s.	1968	300,875.00	300,000	—449.31
37,000	Greyhound Corporation, S. F. Deb. 3s.	1959	37,346.87*	37,740	553.00
200,000	Minnesota Power & Light Co., 1st Mtg. 3 1/8s.	1975	204,508.83*	204,000	3,126.00
100,000	Ohio Power Co., 1st Mtg. 3 1/8s.	1968	101,500.00	106,000	1,622.00
97,000	Oklahoma Natural Gas Co., 1st Mtg. 2 7/8s.	1961	98,154.10*	97,970	1,393.89
50,000	Philadelphia Electric Co., 1st & Ref. Mtg. 2 1/8s.	1978	49,687.50	50,000	—147.75
207,000	Philadelphia Electric Power Co., 1st Mtg. 2 1/8s.	1975	211,742.56*	198,700	2,569.34
200,000	Public Service Co. of Indiana, Inc., 1st Mtg. 3 1/8s.	1975	204,522.57*	203,700	3,126.00
125,000	Puget Sound Power & Light Co., 1st Mtg. 4 1/8s.	1972	129,320.40*	133,750	5,312.50
216,000	Tennessee Gas & Transmission Co., 1st Mtg. Pipe Line 2 1/8s.	1966	218,916.00*	209,520	2,970.00
292,000	United Gas Corp., 1st Mtg. & Coll. Tr. 2 1/8s.	1967	292,000.00	292,000	4,015.00
	Income from bonds called.	110.00
<u>\$2,067,000</u>	Total Public Utility	<u>\$2,106,035.38</u>	<u>\$2,093,710</u>	<u>\$27,954.42</u>
COMMUNICATION BONDS					
\$150,000	American Telephone & Telegraph Co., Conv. Deb. 2 1/8s.	1961	\$155,944.71*	\$162,000	\$1,122.00
150,000	American Telephone & Telegraph Co., Deb. 2 1/8s.	1975	152,531.25*	144,000	2,062.50
200,000	Mountain States Telephone & Telegraph Co., Deb. 3 1/8s.	1978	201,750.00	205,000	2,208.34
52,000	New England Telephone & Telegraph Co., 1st Mtg. 5s.	1952	51,748.00	53,560	2,609.00
100,000	New York Telephone Co., Ref. Mtg. 3 1/8s.	1978	101,430.00	104,000	—17.36
200,000	Pacific Telephone & Telegraph Co., Deb. 3 1/8s.	1978	205,500.00	208,000	—428.80
300,000	Southwestern Bell Telephone Co., Deb. 3 1/8s.	1983	307,500.00	306,000	—260.40
<u>\$1,152,000</u>	Total Communication	<u>\$1,176,403.96</u>	<u>\$1,183,560</u>	<u>\$4,869.60</u>
RAILROAD BONDS					
\$100,000	Chesapeake & Ohio Ry. Co., Gen. Mtg. 4 1/8s.	1992	\$99,464.29	\$127,000	\$2,250.00
75,000	Chicago & W. Indiana R. R. Co., Cons. 4s.	1952	70,357.66	78,000	1,500.00
100,000	Pennsylvania R. R. Co., Cons. Mtg. 4 1/8s.	1960	104,662.50	116,000	2,250.00
<u>\$275,000</u>	Total Railroad	<u>\$274,484.45</u>	<u>\$321,000</u>	<u>\$6,000.00</u>

*After deduction for amortization of premiums on bonds purchased subsequent to January 1, 1940.

SCHEDULE OF SECURITIES OWNED JUNE 30, 1948—Continued

Aggregate par or nominal value	Description	Maturity	Cost, amortized cost, or value at date acquired	Approximate market value	*Net income (8 months)
RAILROAD EQUIPMENT TRUSTS					
\$150,000	Chesapeake & Ohio Ry. Co., Eq. Tr. 2s.	1956-58	\$146,340.34	\$145,500	\$-133.32
300,000	Chicago Burlington & Quincy R. R. Co., Eq. Tr. 2 1/4s.	1958-63	292,507.12	290,000	-75.00
150,000	Pennsylvania R. R. Co., Eq. Tr. 2 3/4s Guar.	1958-63	146,358.96	144,300	-554.15
150,000	Southern Pacific Co., Eq. Tr. 2 1/4s	1956-58	146,251.10	145,500	-203.65
150,000	Southern Railway Co., Eq. Tr. 2 1/4s	1956-58	145,928.69	144,000
\$900,000	Total Railroad Equipment Trusts	\$877,386.21	\$869,300	\$-966.12
INDUSTRIAL AND MISCELLANEOUS BONDS					
\$200,000	American Tobacco Co., Deb. 3s.	1969	\$203,216.26*	\$204,000	\$411.10
240,000	Bristol Myers Co., Deb. 3s.	1968	241,700.00	244,800	-1,000.00
64,000	Devco & Raynolds Co., Inc., S. F. Deb. 2 1/4s	1965	65,785.28*	64,000	3,687.69
195,000	Eastern Gas & Fuel Associates, 1st Mtg. & Coll. Tr. 3 1/4s	1965	195,705.25*	196,978	3,412.50
153,000	Food Machinery Corp., S. F. Deb. 2 1/4s	1962	152,508.18*	151,470	1,268.55
300,000	Goodrich (B. F.) Company, 1st Mtg. 2 1/4s	1965	301,265.18*	300,000	8,250.00
298,000	P. Lorillard Co., Deb. 3s	1963	301,948.24*	303,960	2,579.24
300,000	National Dairy Products Corp., Deb. 2 1/4s	1970	304,618.34*	307,000	5,036.55
400,000	Phillips Petroleum Co., S. F. Deb. 2 1/4s	1964	403,532.30*	404,000	1,091.90
23,000	Pittsburgh Consolidation Coal Co., Deb. 3 1/4s	1965	23,165.51*	125,690	-1,053.95
300,000	Quaker Oats Co., Deb. 2 1/4s	1964	148,922.50	285,000	7,500.00
300,000	Seagram Joseph E. & Sons, Inc., Deb. 2 1/4s	1966	298,500.00	285,000	5,000.00
400,000	Shell Union Oil Corp., Deb. 2 1/4s	1971	403,498.18*	390,000	7,109.36
400,000	Socony-Vacuum Oil Co., Deb. 2 1/4s	1976	390,278.75	376,000	-2,750.71
400,000	Standard Oil Co. of California, Deb. 2 1/4s	1966	410,687.50	388,000	2,091.71
300,000	Swift & Co., Deb. 2 1/4s	1972	301,743.88*	288,000	1,432.41
400,000	Texas Corporation, Deb. 3s	1965	421,698.01*	416,000	6,453.00
250,000	Union Oil Company of California, Deb. 2 1/4s	1970	259,365.85*	247,500	2,623.00
100,000	United States Rubber Co., Deb. 2 1/4s	1976	100,334.97*	94,000	2,419.94
300,000	Westinghouse Electric Corporation, Deb. 2 1/4s	1971	302,162.04*	288,000
\$5,173,000	Total Industrial and Miscellaneous	\$5,240,508.45	\$5,122,398	\$53,342.49
\$19,904,000	BONDS—Funds Invested	\$20,059,296.61	\$20,058,011	\$368,741.11

*After deduction for amortization of premiums on bonds purchased subsequent to January 1, 1940.

SCHEDULE OF SECURITIES OWNED JUNE 30, 1948—Continued

Number of shares	Description	Cost, amortized cost, or value at date acquired	Approximate market value	Net income (8 months)
PREFERRED STOCKS				
1,000	Anchor Hocking Glass Corp., \$4.00 Cum. Pref.	\$112,750.00	\$104,000	\$2,000.00
1,500	Appalachian Electric Power Co., 4½% Cum. Pref.	159,000.00	154,500	5,062.50
2,000	Armstrong Cork Co., \$3.75 Cum. Pref.	205,500.00	196,000	5,625.00
1,500	Bethlehem Steel Corp., 7% Cum. Pref.	183,637.50	207,000	5,250.00
1,477	Bristol Myers Co., 3½% Cum. Pref.	156,300.45	147,700	2,769.38
2,000	Buffalo Niagara Electric Corp., 3.60% Cum. Pref.	207,990.00	170,000	3,600.00
500	Case (J. I.) Co., 7% Cum. Pref.	62,225.00	72,500	1,734.91
600	Cleveland Electric Illuminating Co., \$4.50 Cum. Pref.	68,112.25	66,000	1,350.00
1,000	Columbus & Southern Ohio Electric Co., 4½% Cum. Pref.	115,350.00	98,000	3,190.00
2,000	Consolidated Edison Co. of N. Y., \$5.00 Cum. Pref.	213,490.00	212,000	7,500.00
1,125	Continental Can Co., Inc., \$3.75 Cum. Pref.	115,312.50	109,125	2,109.38
1,145	Corn Products Refining Co., 7% Cum. Pref.	27,183.25	25,085	507.50
1,125	duPont (E. I.) de Nemours & Co., \$4.50 Cum. Pref.	116,125.00	136,125	2,331.26
1,500	Electric Power & Light Co., \$7.00 Cum. 1st Pref.	253,700.00	234,000	5,250.00
1,000	El Paso Natural Gas Co., 4.10% Cum. Pref.	111,442.21	98,000	3,075.00
2,000	General Foods Corp., \$3.50 Cum. Pref.	201,000.00	188,000	5,250.00
1,500	General Motors Corp., \$3.00 Cum. Pref.	187,937.50	184,500	5,625.00
1,000	General Shoe Corp., \$5.00 Cum. Pref.	102,250.00	83,000	1,750.00
1,000	Grant (W. T.) Corp., \$3.50 Cum. Pref.	100,447.91	98,000	1,875.00
1,500	McKesson & Robbins, Inc., \$4.00 Cum. Pref.	144,000.00	138,000	3,000.00
1,000	Northern States Power Co., \$3.60 Cum. Pref.	103,200.00	87,000	1,800.00
1,695	Ohio Power Co., 4½% Cum. Pref.	76,552.00	75,060	2,345.64
1,500	Pacific Telephone and Telegraph Co., 6% Cum. Pref.	235,220.75	211,500	4,500.00
1,000	Panhandle Eastern Pipe Line Co., 4% Cum. Pref.	104,166.68	97,000	2,000.00
1,000	Pillsbury Mills, Inc., \$4.00 Cum. Pref.	107,722.00	102,000	2,000.00
1,000	Public Service Co. of Oklahoma, 4% Cum. Pref.	105,286.00	90,000	2,000.00
2,000	Reynolds (R. J.) Tobacco Co., 3.60% Cum. Pref.	199,683.75	184,000	3,600.00
974	Servel, Inc., \$4.50 Cum. Pref.	109,696.75	97,400	2,191.50
1,054	Sherwin-Williams Co., 4% Cum. Pref.	116,168.60	113,832	3,162.00
1,400	Standard Oil Co. of Ohio, 3½% Cum. Pref. "A"	150,743.69	134,400	2,625.00
250	United States Gypsum Co., 7% Cum. Pref.	45,187.50	44,000	875.00
3,100	U. S. Steel Corp., 7% Cum. Pref.	443,407.57	430,900	16,275.00
Income from stocks called or sold		3,295.00		
40,445	Total Preferred Stocks	\$4,640,788.86	\$4,388,627	\$115,724.07
COMMON STOCKS				
1,900	Abbott Laboratories	\$144,126.20	\$142,500	\$1,650.00
200	Air Reduction Company	11,989.46	4,800	100.00
2,900	American Can Company	257,082.91	243,600	4,950.00
4,400	American Cyanamid Co.	171,180.54	176,000	4,400.00
2,900	American Gas and Electric Company	122,028.40	113,100	5,075.00
2,700	American Telephone & Telegraph Co.	399,025.39	418,500	12,150.00
4,900	Armstrong Cork Company	243,386.48	264,600	9,020.00
4,500	Boston Edison Company	203,214.54	184,500	7,560.00
1,000	Bristol Myers Co.	39,430.33	32,000	1,200.00
2,800	C. I. T. Financial Corporation	141,857.73	134,400	2,800.00
2,600	Chase National Bank of the City of New York	92,769.35	91,000	3,120.00
8,200	Chrysler Corporation	371,763.95	533,000	24,600.00

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SCHEDULE OF SECURITIES OWNED JUNE 30, 1948—Continued

Number of shares	Description	Cost, amortized cost, or value at date acquired	Approximate market value	Net income (8 months)
COMMON STOCKS—Continued				
3,400	Cleveland Electric Illuminating Company.....	\$147,677 29	\$132,600	\$5,440 00
1,300	Coca-Cola Company.....	189,405 75	218,400	3,375 00
2,000	Commercial National Bank and Trust Co. of N. Y.....	86,522 22	80,000	2,000 00
6,356	Commonwealth Edison Company.....	196,326 62	171,612	6,393 30
2,000	Consolidated Edison Company of N. Y.....	67,530 37	64,000	2,400 00
1,000	Consolidated Gas Electric Light and Power Company of Baltimore	90,349 75	50,000	1,800 00
4,300	Continental Can Co.....	172,761 85	159,100	4,300 00
1,200	Continental Illinois National Bank & Trust Co. of Chicago	105,810 00	96,000	2,400 00
7,608	Continental Insurance Co.....	213,933 83	268,356	3,608 00
7,000	Continental Oil Co. of Delaware.....	261,482 12	509,200	14,700 00
5,100	Delaware Power & Light Company.....	105,714 47	81,600	2,550 00
2,000	Dow Chemical Co.....	69,236 04	90,000	1,000 00
2,800	duPont (E. I.) de Nemours & Co.....	454,108 21	520,800	15,000 00
13,100	Eastman Kodak Co.....	458,548 61	563,300	10,350 00
950	Fireman's Fund Insurance Co.....	98,250 00	96,900	375 00
4,788	Food Machinery Corporation.....	162,661 97	191,520	1,992 50
13,200	General Electric Co.....	520,976 06	541,200	8,960 00
7,400	General Foods Corporation.....	309,465 04	288,600	10,000 00
8,000	General Motors Corporation.....	423,115 49	512,000	18,000 00
10,000	Grant (W. T.) Co.....	181,260 49	200,000	10,000 00
400	Guaranty Trust Co. of N. Y.....	98,003 91	109,600	2,400 00
8,500	Gulf Oil Corp.....	379,925 89	671,500	24,968 75
2,000	Hartford Fire Insurance Co.....	179,826 06	222,000	3,008 00
8,500	Humble Oil & Refining Co.....	276,297 45	667,000	20,375 00
3,275	Insurance Company of North America.....	234,197 06	327,500	5,731 25
2,950	International Business Machines Corp.....	219,994 84	430,700	7,000 00
3,500	Johns-Manville Corp.....	131,320 79	179,500	3,375 00
3,100	Kennecott Copper Corporation.....	151,715 82	170,200	4,375 00
4,600	Kresge (S. S.) Company.....	126,662 15	180,200	5,775 00
2,100	Liggett & Myers Tobacco Co.....	189,977 35	180,600	8,400 00
320	Mellon National Bank and Trust Company.....	67,193 07	188,000	1,600 00
2,700	Minneapolis-Honeywell Regulator Co.....	121,115 63	134,200	5,600 00
7,800	Monsanto Chemical Co.....	284,132 58	460,200	9,400 00
6,300	Montgomery Ward & Co.....	339,979 26	378,000	12,000 00
6,900	National Cash Register Co.....	235,308 19	317,400	7,950 00
1,700	National City Bank of New York.....	68,487 50	66,300	1,560 00
1,260	National Fire Insurance Co. of Hartford.....	72,819 72	55,440	1,260 00
1,800	National Union Fire Insurance Co.....	42,656 09	55,800	1,800 00
10,800	Newberry (J. J.) Co.....	143,047 69	356,800	10,800 00
3,400	New Jersey Zinc Co.....	223,962 50	234,600	9,200 00
3,800	Owens-Illinois Glass Co.....	263,802 50	291,400	8,200 00
2,750	Pacific Gas & Electric Company.....	131,073 09	331,500	3,750 00
10,600	Penney (J. C.) Co.....	370,316 50	508,800	13,500 00
1,000	Peoples Gas Light and Coke Company.....	106,350 00	128,100	2,750 00
2,100	Pfizer (Chas.) & Co., Inc.....	121,389 01	128,100	3,150 00
3,500	Philadelphia Electric Company.....	406,048 99	584,000	14,150 00
7,600	Phillips Petroleum Co.....	279,146 50	336,800	6,355 00
8,700	Pittsburgh Plate Glass Co.....	17,135 87	142,800	2,825 00
2,100	Procter & Gamble Co.....	72,455 18	107,800	2,790 00
1,800	Reynolds (R. J.) Tobacco Co., "B".....	97,824 75	107,800	2,790 00
2,200	Scott Paper Co.....	97,824 75	107,800	2,790 00

(Continued on following page)

SCHEDULE OF SECURITIES OWNED JUNE 30, 1948—Continued

Number of shares	Description	Cost, amortized cost, or value at date acquired	Approximate market value	Net income (8 months)
COMMON STOCKS—Continued				
16,400	Sears, Roebuck & Co.	\$374,427 57	\$672,400	\$24,600.00
6,200	Sherwin-Williams Co.	376,082 01	427,800	9,437.50
2,500	Southern California Edison Company, Ltd.	95,449 80	175,000	2,625.00
5,600	Squibb (E. R.) & Sons	140,697 14	151,200	4,200.00
10,700	Standard Oil Co. of Indiana	384,205 57	524,300	15,350.00
6,845	Standard Oil Co. of New Jersey	404,499 59	588,670	17,400.00
1,200	St. Paul Fire and Marine Insurance Co.	90,787 02	92,400	800.00
4,000	Texas Company	209,513 98	252,000	4,350.00
2,700	Timken Roller Bearing Co.	130,378 34	135,000	7,425.00
11,400	Union Carbide & Carbon Corp.	333,875 55	478,800	7,600.00
9,400	United Fruit Company	266,728 02	498,200	18,800.00
4,200	United States Gypsum Co.	368,226 99	457,800	7,875.00
11,400	Westinghouse Electric Corp.	311,102 29	342,000	9,000.00
4,400	Woolworth (F. W.) Co.	193,719 63	211,200	7,200.00
	Income from stocks sold			2,067.40
365,202	Total Common Stocks	\$15,857,291 09	\$19,733,298	\$558,249 20
405,647	COMMON AND PREFERRED STOCKS—Funds Invested	\$20,498,079 95	\$24,121,925	\$673,973 27
	AGGREGATE INVESTMENTS (BONDS AND STOCKS)	\$40,557,376 56	\$44,179,936	\$1,062,714.38*

*Represents total interest and dividend income before deduction of amortization of bond premiums.

SUMMARY OF SECURITY TRANSACTIONS NOVEMBER 1, 1947 TO JUNE 30, 1948

	October 31, 1947—Cash awaiting investment	SALES AND REDEMPTIONS			October 31, 1947—Cash awaiting investment
		Gain	Loss	Book value	
Common stocks		\$32,774.68		\$615,165.09	
Bonds			\$8,433.90	17,593,842.31	
Preferred stocks			7,960.31	235,606.76	
Mortgages		1,234.36			
Sale of stock rights		7,722.82			
Net gain		\$41,731.86	\$16,394.21	\$18,444,614.16	
			25,337.65	25,337.65	
		\$41,731.86	\$41,731.86		
Income applied to amortization of bond premiums					8,519.29
					\$18,729,570.69
Bonds				\$15,783,254.32	
Common stocks				2,754,960.77	
June 30, 1948—Cash awaiting investment					\$191,355.60

REPORT OF THE PRESIDENT
OF THE
CARNEGIE INSTITUTION OF WASHINGTON
FOR THE YEAR ENDING SEPTEMBER 30, 1948

REPORT OF THE PRESIDENT OF THE CARNEGIE INSTITUTION OF WASHINGTON

This annual report, presented to the Trustees as the By-Laws prescribe, finds the Institution engaged in vigorous activity after a valuable year's testing of the departmental programs, the evolution of which after the hiatus of war was described in summary in the report for 1947.

In that report consideration was given both to the way in which programs of research take form, and to the appropriate relationship between the governing board and the scientific staff in institutions of this nature. There has never been any difficulty on this point in the Institution, for the free play of the scientific initiative of the staff is never circumscribed by preconceived notions on the part of the Trustees, or of the President of the Institution for that matter, yet it is well at times to review the relationship.

There is a contrast in this connection that is worth noting. The world is now divided starkly into two groups with vastly different ideologies. These produce utterly different frameworks within which life goes on on the two sides of the iron curtain. This difference extends of course to the fields of science and scientific research. On our side, we hold strongly to the freedom of science, for we believe intensely not only that progress in pursuit of knowledge is made rapidly and securely only when any hypothesis whatever may be entertained by individuals, but also that the survival of hypotheses should be determined only by the rigor of test against cold facts and by the consequent mass of scientific support or denial, untrammelled by any arbitrary rulings from above.

On the other side of the curtain this is not so. In fact, we are told that the philosophy of Mendelism is contrary to the philosophy of Marx and Lenin and must be extirpated, and that those who teach or utilize the genetic laws of inheritance are enemies of the state, to be treated accordingly. It is hard for us to conceive how a body of science in the whole field of biology can be developed under such restraints without becoming ultimately grotesque—a collection of folklore and superstitions rather than of truth. It is hard also to see how if these two systems are in competition there can in the long run be any result, even in the field of the applications of science which is so important for national welfare, other than an outdistancing of the second by the first, an outdistancing which will gradually be so apparent to all as to force a revision in any situation where men's minds are still capable of the ordinary processes of human logic.

To assert as we do that programs of research should arise by reason of the integrated aspirations and judgment of the scientific staff is merely to open a question of some complexity and consequence. We know that in essence the initiation of a research, a quest into the unknown, occurs most fruitfully because of the imaginative power of the qualified mind. We know that the intuitive impulse of the dilettante is no match for the imaginative prospective thinking of a trained and flexible mind. There is before us the example of the giants of research who have

oftentimes made their great successes largely because of their keen judgment as to what was important, possible, and opportune. Yet the evolution of a scientific program in the hands of a qualified scientific staff is more than just this.

In the early days of the Institution great emphasis was placed upon the principle of seeking out the unusual man, and giving him untrammelled opportunity and generous support. The principle is a sound one and it is still in effect. The staff of the Institution, those who have come to the point of acceptance by their colleagues as masters of their art, have indeed been selected by a subtle but nevertheless rigorous process. They do indeed have extraordinary opportunity to pursue their scientific endeavors without interruption by the inconsequential, without entanglement in minutiae of regulations or systems, without the burden of auxiliary duties. They have this opportunity to an extent that is rare, and the results have justified it. Yet the principle must mean much more today than a search for the individual genius. Much as has been accomplished in the past by the lone worker in science, he who neither required anything from his fellows nor gave them much, he who succeeded by his brilliance and his enormous energy; much as is still accomplished at times by men who thus travel alone, the sound progress of a body of science requires even more, and the most valuable scientist in society is that individual who combines with enormous intellectual urge of a personal nature the ability to operate in parallel and in close interchange with colleagues of equal attainments and equal ambitions.

The process by which a group of scientific men evolve a joint integrated program of research is, in fact, impeded by the presence of the obdurate lone worker, and

the Institution in its departments is committed to joint programs. It is always to be hoped that funds of the Institution will be available in the form of grants for the support of others who are working in parallel ways, even though our activities in this regard have necessarily been circumscribed under the pressure of rising costs and reduced rates of income. It is to be hoped also that the individual genius can occasionally be thus found and supported. Some of the brightest pages in the history of the Institution reveal the importance of the Institution's support at an early period to masters working in other organizations.

When we review this history, however, it will be found that the support produced its greatest results ordinarily when it led to progress by an individual who not only could succeed in individual intellectual endeavor of a high order, but also could build a school, or found a branch of science, or gather to him groups of disciples who later permeated the entire field of his interest and furthered it in a dozen centers. The relationship of master and disciple is the simplest form of internal organization for research, and when it is successful it is one of the most salutary forms. This relationship has been prominent in the Institution's activities throughout its history, and we seek to give it larger opportunity as we bring in younger men in greater numbers through our fellowship plans.

Neither the self-realization of the individual genius nor yet the clustering of a coterie at the feet of an individualistic master, however, is of primary importance. Both are of influence in the delicate matter with which we are concerned. Both will affect the development of integrated programs for joint effort. Both can and often do contribute directly and effectively to

the advancement of knowledge. Yet the central question, which remains of first importance, is that of the way in which a group of scientific colleagues of equal stature plan in concert and interrelate their affairs so that out of their combined thinking there comes a group or departmental program such that it not merely will meet the approval of a governing board, but will afford firm basis for the continuing joint effort of qualified minds. In this connection we need to go much farther than merely to use the words collaboration and co-ordination. They are overused in any case, so much so that they are oftentimes utilized merely as an excuse for doing little. Also we need to do more than merely leave the matter to chance or even to good relations such as obtain among the scientists of the Institution.

A fundamental distinction must be kept in mind; there is an essential difference between the conception and the process of an investigation into the unknown. We feel assurance, for example, that Euclid as he launched out on new inquiries acted because of the promptings of his independent mind; yet we know that as he traced his triangles in the sand he correlated and extended the work of earlier geometers. In the process of his investigation, then, in the actual operations by which he achieved new knowledge, he was armed and strengthened, perhaps indeed unwittingly, by the work of predecessors known and unknown. His world of some two millennia ago was a simpler world, where the disciplines were few and each of them was well nigh unitary and independent of the others. Research—the conscious and ordered inquiry into the unknown—then was at best a timid technique. Science—the ordered body of knowledge resulting from research—was barely taking inchoate form. In contrast, ours

is a world in which science and the applications of science through technology and engineering have come to be the dominant factor in life over much of the globe and for the majority of mankind. It is a world, indeed, whose greatest current peril grows out of the clash of opposed philosophies of government expressing antagonistic concepts of how to handle a culture made dynamic by the continual swift change imposed by the unceasing expansion of our knowledge of how to control and to exploit our physical environment. One measure of the change is afforded by the extent to which the process of research—the process as distinguished from the conception—has become a matter of joint effort.

The range of this joint effort is very wide. From the simplest form, that of the relationship of master and disciple which we have already noticed, it extends to the complexity of such vast affairs as the research operations of the Atomic Energy Commission. Between these two extremes are many other forms of organization for joint effort in the conduct of investigations—the university laboratory, the industry laboratory, the endowed research agency of which the Carnegie Institution of Washington is an example.

In the development of departmental research programs within the Institution, this central difference between the conception and the process of research is an important factor. The objectives which the programs seek express the aspirations of the staff; hence at outset the programs are built upon the imaginative prospective thinking of qualified individual minds. But a collection of discrete projects aimed at attaining individual objectives does not constitute a program in the sense in which the departments of the Institution use the term. Though it contemplates specific and

sometimes widely separated objectives determined by individual interests and competence, a program as we conceive it is a synthesis, a summary of joint effort planned for performance. The process as distinguished from the conception of research may be said actually to begin in the preparation of a program thus conceived.

The formulation of such a synthesis is an interesting operation, relying for success on the tempered individualism of a group of colleagues, each of whom, however intense may be his drive toward the goal of his especial choice, recognizes the validity of the goals set by his peers and because of that recognition is prepared to aid them as he hopes to be aided by them. Not only does he evaluate his own purposes and methods in the light of those of his colleagues, but also he is ready to modify his own if necessary to enhance the whole. However clear and specific the particular objective, the investigator of high qualifications can be expected, and expects, to have at his command several possible approaches toward it. In mountain-climbing, there may be one course and no other to gain the peak; in research, the truly adept investigator can discern alternatives. Probably the most important single attribute of the successful scientist is his ability not simply to define the result which he seeks to achieve, but, more than that, to recognize various possible ways of accomplishing it and to devise methods suitable to utilize them.

The core of program-making is in the making of choices among alternative methods in the light of the choices being made by colleagues, so that interrelation and reciprocal support result. When this is well done, the program as a whole is indeed greater than the sum of its parts—the procedures being employed by indi-

vidual investigators or research groups as they work toward the advance of knowledge through the attainment of their particular objectives. The experience of the Institution's departments and divisions in developing programs at the end of the war, in which every scientist in the Institution took part, demonstrated well the validity of this theory. That experience was a good beginning; out of it came for each department an ordered plan for the future, subject of course to change as the art develops, and subject likewise to periodic review, but pointing out objectives, weighing the probable value of planned efforts, and producing a frame of reference against which individual proposals could be balanced. Probably the most important single attribute of a scientific staff is the ability to perform as a group this function of estimating and balancing, integrating and interrelating, which is in essence the intricate means whereby a program comes into being and which in its reflection of the increased interdependence naturally disclosed with the increasing extent of our knowledge emphasizes the fact that the process of research has come to be a matter of joint effort.

The tradition of scientific inquiry and of the laboratory extends so far back in time that we easily overlook the fact that the organized joint effort which we know as research today and which has so much expedited the maturing of science is a fairly recent development. Agencies such as the Carnegie Institution of Washington, endowed directly for the conduct of basic research, are principally creations of the past half century. Aggressive large-scale industrial research—to turn to the applied field—likewise is of comparatively recent origin. Highly effective patterns for the marshaling of joint efforts in basic and in applied research have come into being

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during this period; the pioneering stage in the development of such mechanisms has been passed. The record of growth is encouraging, and it is interesting in its progression through the stages to be expected in new developments. Thus early emphasis was often strong on the individualism of the new enterprise, sometimes indeed so strong as to produce outright separatism. As time went on, there was growing recognition that individuality is not dependent on particularism, and consequently an increasing trend toward collaborative undertakings. As we near the halfway point of the twentieth century, research agencies throughout the country, in fact throughout most of the civilized world, are engaged in probably the largest range of joint activities in history. There is apt illustration in such a concept as Associated Universities, contractor to the Atomic Energy Commission for the operation of the Brookhaven National Laboratory.

The action of the California Institute of Technology and the Carnegie Institution of Washington in inaugurating during the past year a carefully prepared plan for the joint operation of the astronomical observatories on Mount Wilson and Palomar Mountain evidences in yet another way the trend toward joint effort in the process of research. Far more is involved

here than provision for the efficient and economical utilization of the millions of dollars invested in the observatories and their instruments—the greatest concentration of astronomical equipment in the world. Important as it is to facilitate such utilization, the greater significance of the plan lies in two other aspects. One is its formulation of a firm and specific framework for joint action by the two parent organizations—the Institute and the Institution—which though separated by a continent are enabled by this clear statement of common principles to work in effective partnership. The other is the creation by this plan of the entity known as the Mount Wilson and Palomar Observatories and of the Staff of the Mount Wilson and Palomar Observatories. Here, in this Staff which comprises members of the Institute staff and of the Institution staff, we find assembled skilled scientists, amply furnished with the equipment and facilities necessary to their work, and by their unique constitution as a group free to carry out the delicate and all-important function of evaluation and integration fundamental to the unitary research program to be followed. The plan for joint operation thus is first of all a reasoned means to foster the opportunity for joint effort which is fundamental to the process of research.

RESEARCH ACTIVITIES

The reports from the Departments and Divisions of the Institution for the year past present a record of wide variety and interest, and in their general tenor indicate that the mood of coherent vigorous advance which was anticipated from the formulation of research programs following the war has been intensified with time. From the broad range of results reported, certain findings and projected undertak-

ings of particular interest are selected for brief review here.

One of the fields in which astronomical observations have direct and immediate application to terrestrial problems is that of the relation of solar phenomena, such as flares, prominences, and sunspots, to magnetic storms and radio transmission on the earth. For several years the Mount Wilson Observatory has supplied the re-

sults of daily solar observations to the Central Radio Propagation Laboratory at the Bureau of Standards, which correlates them with the results from other observatories and then uses them in the prediction of radio transmission and related phenomena. In a further study of these relations, Dr. S. B. Nicholson in collaboration with Dr. Oliver Wulf of the U. S. Weather Bureau has investigated periodic disturbances of the earth's magnetic field that frequently occur at 27-day intervals, and has been able to correlate them with the first appearance, at successive revolutions of the sun, of long-lived patches of bright flocculi.

The investigation of the general magnetic fields of early-type stars by Dr. Horace Babcock of the Mount Wilson Observatory continues to lead to new and unexpected results that may have important implications for many problems. During the present year the spectrum variable HD 125248 has been found to have a general magnetic field which varies from +7800 gauss to -6500 gauss and back again during each 9-day period of its spectrum variability. The variation of the spectra of stars like HD 125248 has been one of the outstanding mysteries of astronomy, but there are now indications that the solution may be found in terms of the variable magnetic fields that have just been observed. Variations in stellar magnetic fields of the range found by Babcock imply the presence of tremendous electric potentials. This suggests a possible source for the particles with very high energy charges found in cosmic rays.

Some results of marked significance have emerged from the Geophysical Laboratory's investigation of silicates in the presence of water under pressure. New observations by Dr. Morey and his group on the behavior of aqueous mixtures have

demonstrated that the solution of solids in vapor may take place in notable amounts. Water vapor at a temperature of 400° C. and under a pressure of 1000 atmospheres, for example, has been shown to dissolve 7/100 of a gram of sodium disilicate per cubic centimeter. It is now very clear that a gas that is under a high pressure and, therefore, has an appreciable density resembles a liquid rather than a typical gas in respect to its ability to dissolve solids. The solution of solids in gases or vapors is of physical-chemical interest and may represent an important step in some types of mineral deposition.

In another line of experimentation, Dr. Bowen and Dr. Tuttle have obtained detailed information on the conditions under which a series of minerals consisting of hydrated magnesium silicates are formed. For example, the mineral serpentine, which in its fibrous form is one variety of asbestos, can now be readily produced in the presence of water under pressure. It has been shown to have an upper limit of stability of approximately 500° C., higher temperatures causing the serpentine to decompose into forsterite and talc. This limiting temperature of the stable existence of serpentine may turn out to provide one of the most useful of "geologic thermometers" for fixing the temperature of processes that have occurred in certain rocks of the earth's crust.

The Department of Terrestrial Magnetism has been considering two tentative hypotheses on the origin of the earth's main magnetic field. One of these is the so-called "fundamental" theory, recently discussed by Blackett, which assumes that the magnetic field is due to a fundamental property of matter, and is hence associated with the size, mass, and rotation of the earth. On the basis of this idea, a significant portion of the field should remain

without sensible change for hundreds of millions of years. According to another theory the field results from a set of complex phenomena inside the earth's central core, based on known electromagnetic laws. Such a theory might permit large changes and even reversals of the earth's field during millions of years. A critical test of these different explanations might be possible if measurements extending over a sufficiently long period of time were available.

Direct measurements of the direction and intensity of the earth's field date back only a few hundred years. This period is, of course, extremely short by geological standards. The measurements do show, however, that rapid changes are taking place; an apparent decrease of the order of 4 per cent per century in the intensity of the field has been indicated by measurements over large areas of the earth's surface.

The Department's measurements of the magnetization of varves and ocean-bottom samples, carried out by Dr. Johnson, Mr. Torreson, and their colleagues, show that large rates of change can persist only for relatively short epochs. In fact, the current interpretation of these measurements would indicate that the earth's field has been behaving and changing throughout the past million years in much the same way as we now observe, with only modest changes in magnitude and direction. This result is compatible with either the fundamental or the core theory, or both. Efforts are hence being made to extend the measurements, although somewhat roughly, back half a billion years by examination of samples of sedimentary rocks. Meanwhile, the fluctuations in the field noted during shorter intervals of time, after further careful checking, should provide useful new basic data for constructing and testing an

adequate theory of magnetic secular change, such as, for instance, a dynamo theory based on internal circulation of material and heat within the earth's central and supposedly fluid core.

In all investigations of cosmic-ray phenomena, there has been a notable absence of clues leading to the mechanism by which high energy is imparted to cosmic-ray particles, or to the place in the universe where this energy is supplied. Taking account of the deflections of charged cosmic-ray particles by the earth's magnetic field and of the effects of the earth's atmosphere, all direct experimental evidence is consistent with the notion that cosmic rays come at equal rates from all directions. Accordingly, it is even an open question whether cosmic rays originate within or outside our galaxy.

Evidence that some of the cosmic rays may, on very rare occasions, originate on the sun was furnished by the discovery, described in the report of the Department of Terrestrial Magnetism for 1947, of three unusually large and sudden increases in cosmic-ray intensity which occurred during intense chromospheric eruptions in large sunspot groups. The increases were due to particles which had sufficient energy (several billion electron-volts) to reach the earth in middle latitudes and poleward, but insufficient energy to reach the earth at the equator, where they were excluded by the earth's magnetic field. This fact makes it difficult to assume that the cosmic rays responsible for the observed increases originated in the sunspots, because the sun's magnetic dipole (on the basis of Dr. Hale's work at Mount Wilson) is about one hundred million times stronger than that of the earth. This means that a cosmic-ray particle which had insufficient energy to reach the earth's equator could not escape from the sun at solar latitudes

where sunspots occur, unless special means of exit were available.

Preliminary investigations by Mr. Forbush, Dr. Vestine, and Dr. Vallarta, visiting investigator, indicate that such a means may be provided by the combined effects of the permanent magnetic field of the sun and the transient field of the sunspots involved. The field of the spots creates a tunnel through the forbidden regions of Störmer, by which particles may then escape. The existence of the tunnel depends on the relative strength and orientation of the sunspot dipole and that of the sun itself. The energy acquired by the charged particle depends on the time-rate of change of the sunspot magnetic moment. The latter arises from the growth of the magnetic field in sunspots, as observed at Mount Wilson, and from changes in distance between spots. Thus, only when the magnetic moment of the sunspot is large enough, its rate of change adequate, and its orientation favorable will particles be accelerated with cosmic-ray energy and allowed to escape from the sun. This necessary combination of circumstances appears to be rare, though investigation indicates that the circumstances were all favorable at the time of the three unusual increases in cosmic-ray intensity that have been observed.

To determine whether these preliminary conclusions are correct will require the calculation of trajectories of charged particles from the sunspot to determine (1) whether these actually escape from the sun, and (2) in case they do, whether they would reach the earth.

At the Division of Plant Biology, the immediate precursor of chlorophyll, protochlorophyll, has been isolated in pure form and its absorption spectrum has been determined. With this information it is now possible to measure the amount of proto-

chlorophyll present in leaves. It has been found that in barley leaves grown in the dark, which become green when exposed to light at low temperatures, the chlorophyll *a* which is thus formed comes from an equal amount of protochlorophyll.

Some of the separate steps in chlorophyll formation and its photochemical destruction stand out clearly in various genetic mutants of corn. Thus, an albino strain of corn was found which, in the dark, forms as much protochlorophyll as does the normal. Furthermore, investigations by Dr. Smith and Miss Koski show that this albino actually transforms the protochlorophyll into chlorophyll on illumination. The albinism in this particular mutant is due not to a deficiency in chlorophyll formation, but to a rapid destruction of chlorophyll in prolonged illumination. It was possible to force the accumulation of small but definite amounts of chlorophyll in this albino strain by giving short light exposures followed by long dark periods. In contrast with the behavior of this albino strain, some so-called virescent strains were found that are deficient in protochlorophyll production but which transform their small amount of protochlorophyll into chlorophyll and are able to retain it so that they become pale green.

A study of the components of chloroplasts responsible for the evolution of oxygen in photosynthesis has been undertaken by Dr. French and Mr. Milner. Equipment and procedures for the preparation of disintegrated chloroplast material and for the subsequent measurement of its photochemical activity have been devised. The search for suitable techniques for the resolution of disintegrated chloroplasts into fractions consisting of the separate components remains a focus of effort.

The green and yellow pigments of the chloroplasts, center of Dr. Strain's studies,

reflect the origin and development of the plant kingdom. The common nature and the wide distribution of these pigments point to a common origin for the photosynthetic apparatus. Small variations of the pigment systems, as indicated by the occurrence of particular pigments, can be correlated with the phylogenetic development of the major plant families. On the basis of these results, chloroplast pigments provide a significant character for the establishment of plant relationships.

An investigation of uronides as products of photosynthesis is being carried forward by Dr. Spoehr, with the purpose of exploring the possibility that, although plants under most conditions synthesize primarily carbohydrates, the synthesis of other types of compounds is not necessarily excluded.

The range-grass experiments which Drs. Clausen, Keck, and Hiesey are conducting took an unexpected turn a year ago when many offspring of hybrids between species which for the most part reproduce asexually proved to be sexual. Asexual reproduction was a desideratum in these hybrids as a prompt means of perpetuating the desirable ones unchanged. This year it has been found that in the third generation of several of these hybrid populations a large amount of asexual reproduction has been restored. This not only offers the plant breeder a stable hybrid, but permits a wider choice of hybrid combinations than could have been available had each hybrid reproduced asexually from its inception. Consequently, that which at first appeared to be a handicap may actually prove to be a considerable asset.

The grass hybrid progenies of a number of combinations are under test at the three transplant stations of the Institution in California and at nurseries of the U. S. Soil Conservation Service in eastern Washington and southern California.

Some of the most promising of these are now selected for seed increase and further testing by the Soil Conservation Service on a larger scale.

Dr. Chaney's visit to China to examine the recently discovered *Metasequoia*, a living relative of the redwood, which previously was known only as a fossil, established that it is now growing in association with the plants which have been found with it in the fossil state. The continuation in China of the climate which in the Tertiary age was widespread in North America has made possible the survival of this unit of vegetation over a period of scores of millions of years. The fossil floras of the northern hemisphere are today found as a flourishing forest in the sheltered interior of Asia. It is perhaps the most notable case of group continuity through time which is known anywhere in the world. Many of the Tertiary fossils so widespread in the northern hemisphere which had been assigned to *Sequoia* now prove in reality to be *Metasequoia*.

Continued progress was made by the Department of Embryology in studying the development of the human body and in elucidating the physiology of reproduction and gestation in higher mammals and man. Numerous additions were made to the collection of human embryos, and the work of studying, photographing, and modeling the valuable early embryos was actively continued. Advances were made in knowledge of the cyclic changes in the human ovary and in the uterus during the menstrual cycle. The study of the enzyme systems that control the chemical physiology of the embryo is now well under way. A case of quadruplet birth was subjected to thorough embryological analysis.

An important contribution to study of the physiology of human reproduction is Dr. Reynolds' invention of an effective

tokodynamometer, an instrument for recording the contractions of the human uterus during labor so that their efficiency can be measured and the work done can be calculated. Collaboration with the Department of Obstetrics of Johns Hopkins Hospital has already given results of clinical as well as scientific value.

Data that throw new light on our concept of the structure of the unit of heredity—the gene—have been accumulated at the Department of Genetics. Working with maize, Dr. McClintock obtained evidence suggesting that what we have considered to be one gene may in some cases be composed of a number of reduplicate units, and may undergo relatively frequent changes in the number of its components, some of these changes showing up as well defined mutations.

For several years the effect of combination treatments, involving infrared radiation and X-rays, on genetic changes induced in *Drosophila* has been under investigation, and the study has now been extended to include nitrogen mustard. It is known that certain genetic changes can be induced by treatment with either X-rays or nitrogen mustard alone, but not by treatment with infrared. Dr. Kaufmann has found that pretreatment with infrared increases the effect of both X-rays and nitrogen mustard, where posttreatment produces no change. By analysis of the types of hereditary change induced by the combination treatments, he has concluded that infrared pretreatment modifies the materials of the chromosome in such a way as to increase the chances of recombination among the potential breaks induced by ionizing radiations and by nitrogen mustard.

Investigations dealing with the mutation-producing potencies of various chemicals, carried on by Dr. Demerec and his colleagues with *Drosophila* and bacteria,

indicate that mutagenicity is a widespread property of chemicals. That it has not been detected until very recently is probably due to the fact that adequate methods were not available for use in earlier experiments.

Particularly interesting results have accrued from two activities of the Division of Historical Research: Sr. Tejeda's completion of the recording of the remarkable Maya mural paintings at the ruins of Bonampak in Chiapas, Mexico; and the excavation of a very rich early tomb at Kaminaljuyu, Guatemala, by Mr. Shook.

The Bonampak paintings, covering the walls of a small three-room temple, vividly illustrate Maya ceremonial practices and methods of warfare during the Classic Period, more than a thousand years ago; they also reveal, more strikingly than has any previous find, the high development by the ancient Maya of this form of art. The photographing and copying in water color of the frescoes of one room were accomplished in 1947. In 1948 those of the second and third rooms were copied. In the second room are depicted with extraordinary vigor and naturalism scenes of battle and the treatment of captives; in the third, elaborately costumed figures are shown taking part in a religious observance. Both Bonampak expeditions were carried out in co-operation with the Mexican government and were financed by the United Fruit Company.

The Kaminaljuyu tomb, discovered in the course of brickmaking operations, which were halted by the Guatemala government to permit careful excavation of it, contained the skeleton of a priest or ruler, accompanied by lavish funeral offerings of pottery, carved stone vessels, and jade ornaments. Its significance, aside from the intrinsic beauty of these objects,

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lies in the fact that the interment was made at about the beginning of the Christian era. It indicates much greater tech-

nological progress and a far more sharply stratified society than were believed to have been attained at so early a date.

STAFF

I have with regret to record the loss to the Institution through death of two of its senior scientists: Dr. George L. Streeter, Director of the Department of Embryology from 1918 to 1940 and Research Associate in that Department thereafter, who died July 27, 1948; and Dr. Sylvanus G. Morley, a leader in the Institution's archaeological researches since 1914, whose death occurred on September 2, 1948.

Dr. Streeter came to the Institution in 1914 from the University of Michigan, where he had been professor of anatomy. Succeeding to the Directorship of the Department of Embryology after the untimely death of Dr. Franklin P. Mall, he carried on the work as planned by the founder of the Department, but with a devotion to the science and with a perfection in technical methods that were characteristically his own. He published a long series of researches on the form, general anatomy, and measurements of the human embryo, on the development of the central nervous system and of the ear, and on embryonic abnormalities. As editor from 1917 through 1940 of the Department's "Contributions to Embryology," Dr. Streeter made the series famous among the world's archives of science. With the aid of colleagues and technical assistants largely inspired and trained by himself, he built up the collection of embryos begun by Dr. Mall to its present pre-eminent place, and he was in his later years generally recognized as the world's foremost specialist on human embryology.

During the period 1935-1940, when the affairs of the biological departments of

the Carnegie Institution were to a certain extent administered jointly, Dr. Streeter served as chairman of the division of animal biology. At this time he was called on also to assist in the planning of the addition to the Administration Building on P Street, taking an especial interest in the design of the Elihu Root auditorium. In these and many other duties during his long period as a Director, Dr. Streeter faithfully and effectively served the Institution as well as his Department and the field of science to which his career was devoted.

Dr. Morley became a Research Associate of the Institution in 1914, an Associate in 1918. During his early years with the Institution he carried out a long series of most difficult explorations in the tropical jungles of southern Mexico and northern Central America, which resulted in more than doubling the previously known number of Maya hieroglyphic inscriptions. He served in the Office of Naval Intelligence during World War I, and shortly after its conclusion organized a program for intensive study of the Maya, with excavations at Chichen Itza in Yucatan, Uaxactun in Guatemala, and Copan in Honduras, as well as collateral researches in documentary history, ethnology, linguistics, and other pertinent fields. Another important aspect of the program which he led was the repair and stabilization of excavated structures and the re-erection and protection of carved stone monuments, to preserve for future study and observation many of the finest examples of Maya architectural and sculptural art. Dr. Morley's tireless energy and his unbounded

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enthusiasm for all things Maya resulted in many fundamentally important contributions by himself; in promoting and stimulating the efforts of others; and, by his lectures and writings, in arousing wide popular understanding of and interest in that most remarkable of pre-Columbian New World civilizations.

During the past year, recognition of the contributions of Institution people—Trustees, administration, and staff—to the war effort beyond those noted in the pamphlet of record published in 1946 has brought honors in medals, appointments, and cer-

tificates of appreciation to more than a score of individuals. These have duly been reported to the Trustees through the records of the Executive Committee.

To Dr. Merle A. Tuve, Director of the Department of Terrestrial Magnetism, in recognition of his outstanding scientific service in such diverse fields as the study of interaction between nuclear particles and the development of the proximity fuze, the Research Corporation Award for 1947 was presented at a dinner given in his honor in Washington on November 19, 1947.

FINANCES

In accordance with the change of the fiscal year authorized by amendment of the By-Laws on December 12, 1947, this report covers a period of eight months ending June 30, 1948. Because of the change, it was necessary to make use of reserves by transfer of a moderate amount to supplement income collections for that period. This was anticipated when the fiscal change was proposed, and appropriate provisions were authorized to meet the situation. Now that the transition has been accomplished, we may expect to operate during the next fiscal year at an effective level within the funds estimated

to be available. Experience gained during the short period of operation under the revised procedure clarifies the preparation of future budget proposals.

During the fiscal period just ended there has been prepared a proposed Manual of Fiscal Procedures, which after review by the Finance Committee and the Executive Committee will be brought before the Board of Trustees. The study which has been made was based on consideration of the fiscal actions of the Institution since the time of its founding, and the adoption of the Manual will simplify and facilitate our operations.

GORDON S. RENTSCHLER

The death, on March 3, 1948, of Gordon S. Rentschler was a profound loss not only to the world of finance, in which he had established a record notable for clarity of vision and integrity of purpose, but also to the world of education and research, to which as life trustee of Princeton University and of the Massachusetts Institute of Technology he had been a counselor of repeatedly proved acumen and insight. None regarded more seriously than Mr.

Rentschler the responsibilities involved in such service to institutions in the academic world, and it was therefore a heartening and significant event when in 1946 he accepted the added duties of membership on the Board of Trustees of the Carnegie Institution of Washington. Even in the short period of his service he brought markedly valuable wisdom to the conduct of our affairs, and it is with a sense of deep loss that we reflect upon his passing.

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RICHARD P. STRONG

Dr. Richard P. Strong, internationally known authority on tropical diseases and epidemics, died on July 4, 1948. A Trustee of the Carnegie Institution of Washington from 1934 until his death, Dr. Strong had aided the Institution with particular effectiveness through his service as a member of the Trustees' Standing Committee on Biological Sciences, to whose work he

was able to bring valuable firsthand knowledge in matters biological and medical throughout the world. The Richard Pearson Strong medal for distinguished achievement in tropical medicine, established in 1944 by the American Foundation for Tropical Medicine, is but one of many reminders of his vigorous and creative service to medical science.

JOHN J. PERSHING

General of the Armies John J. Pershing, whose distinguished career had included service as a Trustee of the Carnegie Institution of Washington from 1930 to 1943, died on July 15, 1948. From the wide range

of his experience of men and great issues, General Pershing contributed generously in the formation of broad policies of the Institution during his incumbency as a member of the Board.

REPORTS OF DEPARTMENTAL ACTIVITIES AND CO-OPERATIVE STUDIES

ASTRONOMY

Mount Wilson and Palomar Observatories

TERRESTRIAL SCIENCES

Geophysical Laboratory

Department of Terrestrial Magnetism

Special Projects

BIOLOGICAL SCIENCES

Division of Plant Biology

Department of Embryology

Department of Genetics

Special Projects

HISTORICAL RESEARCH

Division of Historical Research

Special Projects

MOUNT WILSON AND PALOMAR OBSERVATORIES

OPERATED BY THE CARNEGIE INSTITUTION OF WASHINGTON
AND THE CALIFORNIA INSTITUTE OF TECHNOLOGY

Pasadena, California

IRA S. BOWEN, *Director*

OBSERVATORY COMMITTEE

IRA S. BOWEN, *Chairman*
WALTER S. ADAMS
EDWIN P. HUBBLE

MAX MASON
RICHARD C. TOLMAN
E. C. WATSON

As announced in 1945, an agreement was reached during that year between the Carnegie Institution of Washington and the California Institute of Technology for the joint operation of the Mount Wilson Observatory and the Palomar Observatory. The detailed plans for this operation were formulated and approved by the two institutions during the present report year, and the plans became effective on April 1, 1948. The official statement of this plan follows.

UNIFIED OPERATION OF THE MOUNT WILSON AND PALOMAR OBSERVATORIES

The Palomar Observatory of the California Institute of Technology and the Mount Wilson Observatory of the Carnegie Institution of Washington constitute a unique and unprecedented concentration of scientific facilities in astronomy. Outstanding scientific talent is present both in the field of astronomy and in the neighboring fields of physics, mathematics, and chemistry. Hence extraordinary opportunities for accomplishment exist. For some time the Institute and the Institution have recognized the advantages implicit in the creation of a great astronomical center in which a unitary scientific program would be pursued under highly favorable circumstances, which would attract distinguished investigators to collaborate with the staff of the observatories in scientific mat-

ters, and which would draw young men of great ability to graduate studies where they might enjoy the inspiration of leading minds, and familiarize themselves with powerful tools of exploration. For this purpose a unified scientific program is planned in which the observatories will function as a single scientific organization. Under the following plan of unified operation, approved by the Board of Trustees of the Carnegie Institution of Washington on December 12, 1947, and by the Board of Trustees of the California Institute of Technology on January 26, 1948, all of the equipment and facilities of both observatories are made available for the astronomical investigations of the staff members of the combined observatories and collaboration in furthering graduate study in a Department of Astronomy is undertaken.

I. Name

The name of the combined observatories will be the Mount Wilson and Palomar Observatories. This name, with the names of the two supporting institutions, shall appear on letterheads and on all publications by the staff members of the Observatories.

II. Administration

The two observatories shall be placed under the administration of a Director. An Observatory Committee of six members advisory to the Director, and including him as one of the six members and as Chairman, shall be

appointed. Three members of the Observatory Committee, including the Director, shall be appointed by the Carnegie Institution, and three members by the California Institute. Appointments by one institution shall be subject to approval by the President of the other.

III. *Research Program*

There shall be a unitary scientific program of the Mount Wilson and Palomar Observatories. It shall comprise investigations deriving from the judgment of the scientific staff, as studied and formulated by a research committee. The program shall be subject to review and to modification as the advance of knowledge indicates.

IV. *Educational Program*

A department of astronomy giving instruction in this subject will be developed by the California Institute as part of its present Division of Physics, Astronomy, Mathematics, and Electrical Engineering. The Chairman of this Division will be in charge of educational policies of the department and of the supervision of its students. He will normally be one of the California Institute members of the Observatory Committee and this Committee will be advisory to him on the educational problems of the department. It is expected that members of the staff of the Observatories, including those whose salaries are paid by the Carnegie Institution, will participate in the instructional program of the department of astronomy as opportunities for presentation of specialized subjects appear, to which they are willing and able to contribute. Members of the staff of the Observatories who participate in the instructional program will become members of the Division of Physics, Astronomy, Mathematics, and Electrical Engineering.

V. *Staff*

There shall be a scientific staff known as the Staff of the Mount Wilson and Palomar Observatories, the members of which shall

be those members of the scientific staffs of the Carnegie Institution and the California Institute participating in the unitary scientific program of the Observatories. Each member shall have the official title of Staff Member of the Mount Wilson and Palomar Observatories, and his appointment to the Staff of the Observatories shall be recommended by the Director, after consultation with the Observatory Committee, to the Presidents of the Carnegie Institution and the California Institute.

VI. *Guest Investigators*

To increase the scope of the work, guest investigators from other institutions will be invited by the Director, after consultation with the Observatory Committee, to make use of the facilities of the Observatories in collaboration with the Staff, and in such manner as he considers will best advance the scientific program of the combined observatories. In general, these invitations will be issued after presentation of a program of proposed studies by the guest investigator.

Except in special cases the Observatories will not expect to assume financial responsibility for the payment of salaries or grants to guest investigators, since such will usually be available from the organization of which the guest investigator is a member. Such organization should feel free to publish the results of studies pursued by its guest investigator, making proper acknowledgment of the aid rendered by the Observatories through use of facilities and otherwise.

VII. *Budget Procedures*

An estimate of the operating expenses required to maintain the activities of the Mount Wilson and Palomar Observatories shall be made up annually by the Director after consultation with the Observatory Committee. This estimate shall be divided into two parts, the one chargeable to the budget of the Carnegie Institution and the other to that of the California Institute. Insofar as practicable, operating and capital expenses of the Mount Wilson Observatory and the Santa Barbara

Street building will be assigned to the Carnegie Institution budget, and the operating and capital expenses of Palomar Observatory and the Robinson Astrophysical Laboratory to the California Institute budget. Salaries of staff members of the Carnegie Institution and the California Institute who are also members of the Staff of the Mount Wilson and Palomar Observatories will be carried in the budgets of the respective institutions. Each budget will be submitted for approval to the Trustees of the appropriate institution through the usual channels, at which time each institution will be informed of the budget proposed by the Director to the other institution.

Requisitions against the approved budgets will be drawn by the Director, and will be processed and paid for through the purchasing and accounting offices of the appropriate institution. Equipment purchased with the funds of a given institution will remain the property of that institution.

In general each institution will utilize for the combined purpose funds already available for expenditure in this general field. Recognizing the opportunities which may reside in expansion of effort, in case either institution should seek further funds for such utilization it will be aided in such search in every practicable way by the other institution.

VIII. *Shops and Other Service Facilities*

The use of various service facilities of the two observatories, such as machine shops, optical shops, photographic departments, design departments, and so forth, will be scheduled by the Director after consultation with the Observatory Committee in the manner which seems desirable to provide for most efficient operation.

IX. *Continuity of Unified Operation*

It is expected that the unified operation of the Observatories will be continuing and, indeed, strengthened with the passage of time and the realization of the expected benefits. Should an unforeseen situation arise in the future when it seems best that any part or

all of either the Institute's or the Institution's work should be carried forward independently of the unified operation, this may be given effect by notification of one to the other not less than one year in advance of the proposed action.

As now constituted, the operating personnel of the Mount Wilson and Palomar Observatories consists of the former staff of the Mount Wilson Observatory, personnel necessary to maintain and operate the physical plant on Palomar Mountain, and certain members of the staff of the California Institute who will carry on investigations at the Observatories. At the end of the report year the permanent members of the former Mount Wilson Observatory staff number 47, of whom 15 are astronomers. A staff of approximately 15 will be required to operate the facilities on Palomar Mountain and to provide the supporting shop and secretarial work in Pasadena. It is expected that three or four members of the California Institute staff will participate actively in observations during the first year of joint operation.

A very important step in the development of the Mount Wilson and Palomar Observatories is, of course, the impending completion of the major new equipment on Palomar Mountain. After very extensive tests in Pasadena in the fall of 1947 showed that a satisfactory figure had been achieved, the 200-inch mirror was transported to the dome on Palomar Mountain on November 18 and 19. The dome and the essential mechanical parts of the telescope had been completed prior to this date. On arrival at the dome, the mirror was placed directly in the aluminizing chamber and given its first coat of aluminum. As soon as the necessary adjustments could be made, the mirror was then mounted in its permanent position in the telescope.

The first visual tests of the telescope were made on December 21, and the following night photographic plates were exposed using a full-size Hartmann screen.

Since then an extensive series of tests has been carried out to study the behavior of the mirror and the effectiveness of the support system in maintaining a constant figure as the telescope is pointed toward various positions in the sky. Any mirror of this size is of necessity very much more flexible than the mirrors normally used in smaller telescopes. It is therefore essential to have a more elaborate support system, capable of applying the proper supporting forces with a high degree of precision in all orientations of the mirror.

The present support system was designed and built in 1935, and the mirror has been mounted on it throughout the grinding and figuring process. The early laboratory tests of the mirror had raised the suspicion that the friction of certain members of the support system was too great to permit the system to apply the proper balancing forces with the required accuracy. The more systematic measures of the figure of the mirror which were possible with the mirror in place in the telescope confirmed these fears. The part of the support system containing these members has now been redesigned and a pilot model built. As tests of this pilot model indicate that a very substantial improvement has been attained in the new design, a complete set of parts has been ordered. It is anticipated that these parts will be completed and installed and that the mirror will be ready for further tests not later than October 1, 1948.

Early in the tests of the telescope with the mirror in place, a serious vibration of the telescope about the polar axis developed while the telescope was being driven to follow a star. After some delay this was eliminated by an adjustment and a slight modification of the drive mechanism. Ex-

cept for this vibration, the drive and control mechanisms have functioned very satisfactorily.

In the design of the telescope tube great care had been taken to reduce flexure to a minimum as the telescope is given different orientations. Furthermore, the residual flexures that could not be completely eliminated had been adjusted to compensate each other in such a way as to reduce as far as practicable the motion of the optic axis of the main mirror with respect to the photographic plate. Tests of the telescope have shown that this was achieved to a remarkable degree, since in no orientation of the telescope does the intersection of the optic axis with the photographic plate depart by more than 0.01 inch from its mean position. This will render completely practicable the use of Ross correcting lenses at the prime focus to enlarge the field of the telescope.

While these tests and adjustments of the main instrument have been in progress, the engineering staff and shops have been proceeding with the design and construction of the very large amount of auxiliary equipment, such as spectrographs, that will be required before the 200-inch telescope will be ready to carry out a well rounded program of astronomical observations.

The mounting for the 48-inch Schmidt camera was completed this year, and extensive tests and adjustments of the drive mechanism have been carried out. The optical parts have also been completed and will probably be installed in August or September 1948.

Formal dedication ceremonies for the Palomar Observatory were held on June 3, 1948, in the dome of the 200-inch telescope. These were followed on July 1 by a scientific session on the mechanical and optical features of the telescope and the program planned for it. This session was held at the Observatory in connection with a joint

meeting of the American Astronomical Society and the Astronomical Society of the Pacific.

At the formal dedication ceremonies, announcement was made of the unanimous adoption by the Board of Trustees of the California Institute of Technology on May 10, 1948, of the following resolution:

The Board of Trustees of the California Institute of Technology hereby resolve that the 200-inch telescope of the Palomar Mountain Observatory shall hereafter be known as

THE HALE TELESCOPE.

By this action the Board of Trustees seeks to

recognize the great achievements of Dr. George Ellery Hale (1868–1938) who served as Director of the Mount Wilson Observatory from 1904 to 1923, who served as a member of the Board of Trustees of the California Institute from 1907 to 1938, who originated the bold conception of the 200-inch telescope and whose brilliant leadership made possible its design and construction. As this great instrument probes the secrets of the universe, it is fitting that it should stand also in memory of the great scientist and the great leader who contributed so brilliantly to the science of astronomy and who served so ably his community and his nation.

STAFF AND ORGANIZATION

RESEARCH DIVISION

Solar Physics: Harold D. Babcock, Seth B. Nicholson, Joseph O. Hickox, Edison R. Hoge, Edison Pettit, Robert S. Richardson, Mary F. Coffeen, Myrtle L. Richmond, Irene Whitney.

Stellar Spectroscopy and Motions: Paul W. Merrill, Horace W. Babcock, Ira S. Bowen, Jesse L. Greenstein, Milton L. Humason, Alfred H. Joy, Rudolph Minkowski, Roscoe F. Sanford, Olin C. Wilson, Ralph E. Wilson, Fritz Zwicky, Sylvia Burd, Cora G. Burwell, Mary F. Coffeen, A. Louise Lowen, Barbara Olsen.

Nebular Photography, Photometry, and Spectroscopy: Edwin P. Hubble, Walter Baade, Milton L. Humason, Joseph J. Johnson, Rudolph Minkowski, Edison Pettit, Albert G. Wilson, Fritz Zwicky, Alice S. Beach.

Physical Laboratory: Robert B. King.

Editorial Division: Paul W. Merrill, editor; Elizabeth Connor, assistant editor and librarian; Helen Stecki Czaplicki, secretary.

Alfred H. Joy has continued as secretary of the Observatory throughout the year.

Mr. Harold D. Babcock retired on February 1, 1948, after thirty-nine years of service at the Observatory. Mr. Babcock made some of the earliest spectroscopic

observations with the 60-inch telescope. He then turned his attention to laboratory and solar problems and made major contributions to the determination of standards of wave length and to studies of the spectrum of the night sky and of the Zeeman effect. He has also made exhaustive studies of the general magnetic field of the sun. He collaborated in the *Revision of Rowland's preliminary table of solar spectrum wave lengths* and more recently has extended this table greatly in both the infrared and the ultraviolet.

Mr. Babcock has been asked to continue as part-time consultant on ruling-engine problems for one year after his retirement from full-time duties at the Observatory.

Effective June 30, 1948, Dr. Robert B. King transferred from the Observatory to the staff of the physics department of the California Institute. It is anticipated that in his new position he will continue his very important studies of transition probabilities. This transfer brings to a close the active work of the physics laboratory of the Mount Wilson Observatory, which has played such a large role in the programs of the Observatory. This discon-

tinuance of the physics laboratory as a part of the Observatory operations is made possible by the close co-operation with all departments of the California Institute which is provided by the plan for the joint operation of the Mount Wilson and Palomar Observatories. In the future the large and well equipped physics department of the California Institute will be available to fill the role played by the former physics laboratory of the Observatory.

Miss Myrtle Richmond retired from the computing staff on October 1, 1947. She had been connected with the Observatory since 1913 and has ably assisted in a large number of solar and stellar investigations.

GUEST INVESTIGATORS

Quantitative photometric measurements on interstellar lines were made by Dr. Lyman Spitzer, of Princeton University Observatory, in the summer of 1947. Equivalent widths were obtained for distant reddened stars at longitudes where the radial velocity due to galactic rotation is small, and also for distant unreddened stars. Analysis of these observations, together with the previous data of Merrill, Sanford, O. C. Wilson, and Burwell, indicates that interstellar sodium vapor is apparently concentrated in the same clouds which produce interstellar reddening. In particular, the correlation of the doublet ratio D_2/D_1 with the color excess E_1 , for stars showing the same average equivalent width $D_1 + D_2$, is confirmation of this belief. The analysis also indicates that thin, relatively transparent clouds and thick, relatively opaque clouds must both be assumed to be present.

In addition, microphotometric tracings of K and H were made from about half the plates taken by Adams in his survey of interstellar calcium. These are now being reduced to provide accurate information both as to the equivalent widths of K and

H and as to the profiles of the individual components in each line. High-dispersion spectrograms were taken of the D lines in stars showing several strong, widely separated components of K and H. Several of these show moderately strong components of the D lines also. It is planned to obtain additional data on D-line components.

During a short visit in August 1947, Dr. A. E. Whitford, of Washburn Observatory, used a lead sulfide photoconductive cell to investigate the infrared radiation of stars, extending the study out to 2.1 microns. Comparison of space-reddened and normal B stars showed that the interstellar-reddening curve has a reverse curvature in the region beyond 1 micron, as had been predicted theoretically by Oort and van de Hulst. This result narrows the uncertainty about the total absorption correction to be applied for a given observed reddening. The reverse curvature reduces the residual absorption at 2 microns to a relatively negligible amount, and strengthens the hope that infrared surveys may show objects previously hidden by the dust clouds of the Milky Way.

During the fall of 1947, Dr. Erik Holmberg, of Lund Observatory, carried out studies of the distribution of luminosity and color in certain spiral nebulae having suitable inclinations to the line of sight and in members of the local group. A total of 150 plates were taken with the 60-inch telescope and 100 with the 10-inch. Appropriate plates and filters were used to give both photographic and photovisual magnitudes, and all nebular exposures were calibrated by means of the north polar scale, the polar stars being photographed out of focus. These plates will be used as the basis of an investigation of the distribution of absolute magnitudes of the nebulae and for the discussion of a number of other problems.

Dr. C. G. Abbot and Mr. L. B. Aldrich, of the Smithsonian Institution, continued tests of an improved radiometer at the 100-inch telescope and made preliminary measurements of the energy distribution in the spectrum of several of the brighter stars.

Dr. Martin Schwarzschild, of Princeton University Observatory, in collaboration with Dr. Adams has completed his investigation of Eta Aquilae, using high-dispersion coude spectra. In his study of period changes of short-period variables, 20 new blue plates and 14 new red plates of Messier 3 were taken during the spring of 1948 to supplement the plates taken in 1946.

A new investigation was started by Schwarzschild regarding the possible difference in chemical composition between stars of Baade's population I and those of population II. The necessary spectra of 9 dwarfs of type F (5 with low velocities and 4 with high velocities) were taken with the 100-inch coude spectrograph. The reduction of the photometer tracings of these spectra will be undertaken at Princeton University. In collaboration with Richardson, Schwarzschild has undertaken a program of investigation regarding the possible existence of a noise zone in the interior of red giants. The corresponding models have been computed for representative cases.

In continuation of work done in previous years, Mr. W. C. Miller and Dr. A. G. Mowbray, both of Pasadena, have obtained many slit spectrograms with the large telescopes, mainly of faint recently

discovered Be stars. In addition, Mr. Miller has taken 32 objective-prism photographs with the 10-inch telescope.

INSTRUMENT DESIGN AND CONSTRUCTION

Design: Edgar C. Nichols, chief designer; Harold S. Kinney, draftsman.

Optical Shop: Don O. Hendrix, superintendent; Floyd Day, optician.

Instrument Shop: Albert McIntire, superintendent; Elmer Prall, instrument maker; Fred Scherff, Oscar Swanson, Albert Labrow, Donald Yeager, machinists; Harry S. Fehr, cabinet maker.

MAINTENANCE AND OPERATION

Office: Anne McConnell, bookkeeper; Dorothea Neuens, stenographer and telephone operator.

Operation: Ashel N. Beebe, superintendent of construction; Kenneth de Huff, engineer; Murdoch McKenzie, janitor and relief engineer; Thomas A. Nelson, Ralph Bennewitz, Eugene Hancock, night assistants; Emerson W. Hartong, truck driver and machinist helper; Anthony Wausnock, Margie Wausnock, Alexander Kochanski, stewards; Arnold T. Ratzlaff, Homer N. Joy, janitors.

As indicated above, the major telescopes on Palomar Mountain were not ready for operation at the end of the report year. As a result, the staff of instrument design and construction and of maintenance and operation for the Palomar Mountain part of the Observatories has not been organized on a permanent basis. This part of the staff of the Observatories has therefore been omitted from the above lists.

OBSERVING CONDITIONS

The season of 1947-1948 has the distinction of having had the lowest precipitation (19.15 inches) in the 44 years of records at the Observatory on Mount

Wilson, the second lowest having been the season of 1923-1924 with a total of 19.40. The months of December and February each had only 6 totally cloudy

days; January had no day that was totally cloudy, and only 4 that were partly cloudy. The total snowfall was 44 inches.

Solar observations were made on 322

days between July 1, 1947 and June 30, 1948. The 60-inch telescope was used on 281 nights and the 100-inch telescope 300 nights during this period.

SOLAR RESEARCH

SOLAR PHOTOGRAPHY

Solar photographs were made on 322 days between July 1, 1947 and June 30, 1948, by Hickox, Hoge, Nicholson, and Richardson. The numbers of photographs of various kinds were:

Direct photographs	644
<i>Hα</i> spectroheliograms of spot groups, 60-foot focus	980
<i>Hα</i> spectroheliograms, 18-foot focus	1,244
K2 spectroheliograms, 7-foot focus	13,370
K2 spectroheliograms, 18-foot focus	939
K prominences, 18-foot focus	1,245

SUNSPOT ACTIVITY

The magnetic classification and study of sunspots and related phenomena have been continued by Nicholson and Miss Whitney. Co-operative programs have been carried out with the United States Naval Observatory, the Observatory at Kodai-kanal, the Department of Terrestrial Magnetism of the Carnegie Institution, and the Central Radio Propagation Laboratory of the National Bureau of Standards.

During the calendar year 1947, observations were made on 337 days, none of which was without spots. The total number of sunspot groups observed was 663 compared with 416 in 1946. The number of groups in the northern hemisphere increased from 200 in 1946 to 309 in 1947; in the southern hemisphere, from 216 to 354.

The monthly means of the number of groups observed daily during the past two and one-half years are given in the accompanying table.

Month	Daily number		
	1946	1947	1948
January.....	4.2	9.9	10.6
February.....	7.7	10.5	8.1
March.....	6.7	11.8	8.0
April.....	7.1	11.5	14.4
May.....	7.2	16.8	12.7
June.....	7.3	13.4	12.2
July.....	8.2	12.9
August.....	8.5	14.1
September.....	8.9	13.7
October.....	9.8	12.4
November.....	9.8	10.8
December.....	8.5	10.1
Yearly average.....	7.8	12.3

The number of sunspot groups observed daily decreased from the high maximum of 16.8 in May 1947 to less than half that number in February and March 1948. This rapid decrease, however, was not due primarily to changes in the eleven-year cycle, but to one of the irregular short-period fluctuations characteristic of sunspot variations. After the low activity of March, that of April was very high, second in this cycle only to that of May 1947. Since the peak in the smoothed sunspot curve for 1948 was a little lower than the similar peak for 1947, it is probable that the maximum in 1947 will be the maximum for the cycle.

SUNSPOT POLARITIES

Magnetic polarities in each spot group have, so far as possible, been observed at

least once. The classification of groups observed between July 1, 1946 and June 30, 1947 is indicated in the accompanying table. "Regular" groups in the northern hemisphere are those in which the preceding spot has S (south-seeking) polarity and the following spot N polarity; in the southern hemisphere the polarities are reversed.

Hemisphere	Regular	Irregular	Unclassified
North.....	221	4	51
South.....	254	8	69
Whole sun	475	12	120

FLARES

As a result of the discovery of apparent relations between solar flares and radio transmission, terrestrial magnetism, and cosmic rays, there has been a steadily increasing interest in data on flares and the spot groups associated with them. Although the positions and intensities of flares are published in the *Quarterly Bulletin on Solar Activity* of the International Astronomical Union, it is often desirable to have in addition information on the type of spot group involved in the flare and the distribution of the bright material, as well as the position of other spot groups upon the disk at the time the flare occurred.

To supply this need, tracings have been taken by Richardson from the 17-inch drawings of spot groups made daily at the 150-foot sun-tower telescope, showing all spot groups visible upon the disk on days when flares were observed at Mount Wilson. A sketch was then made upon the tracing showing the flare as it appeared on the spectroheliograms. Diagrams of the sun are now available for quick reference showing all flares observed at Mount Wil-

son from 1917 to 1947, inclusive. A statistical study of this material is in progress.

PROMINENCES

The interactive prominence of September–October 1947, which was one of the few showing measurable motions in the connecting streamers, has been studied by Pettit. Plates taken with the 13-foot spectroheliograph showed knots in two parallel streamers moving in opposite directions. This would seem to indicate the exchange of gases in the two component prominences with mean velocities of 80 km/sec. This prominence was followed for a period of 4 days; the final stage was quasi-eruptive or eruptive.

SOLAR SPECTROSCOPY

H. D. Babcock, Charlotte E. Moore, and Mary F. Coffeen have published a revised description of the solar disk spectrum in the range $\lambda 2935$ to $\lambda 3060$. All the observations were made at the Hale Solar Laboratory in Pasadena with second- and third-order spectra from a 21-foot concave grating. Wave lengths on the International scale and visual estimates of intensity are given for 665 solar lines. The new intensities are consistent with those found in the region of greater wave lengths, thus removing some long-standing anomalies in Rowland's *Table*. Three-fourths of the tabulated lines are identified. Spectra of metals in the singly ionized state are prominent.

The red bands of atmospheric oxygen. H. D. Babcock and Dr. Luise Herzberg, of Yerkes Observatory, have discussed the structure of the visible bands of oxygen, long familiar in superposition on spectra of celestial objects. All three isotopic forms of the molecule were studied, and, by analysis of precise measurements of wave length, revised values of the molecular

constants were found having higher accuracy than is known for any other molecule. The results have been made available to chemists and physicists in several laboratories, where immediate applications are being made to such diverse problems as the atmospheric absorption of microradio-waves and the computation of the entropy of oxygen.

GENERAL MAGNETIC FIELD OF THE SUN

Six series of measurements with the Lummer plate, made in 1940, 1941, 1943, 1946, and 1947 by H. D. Babcock, have been summarized. Roughly one-half of the measurements indicate a general field of polar intensity of from 5 to 60 gauss, and the remainder show nothing significant. Until additional measurements are completed, a provisional conclusion is that Hale's results are supported and that, as he surmised, the sun's field is variable.

SOLAR ABSORPTION COEFFICIENTS

In continuation of the study of the distribution of violet light across the sun's disk, drift curves were made on all clear days until January 1948 by Nicholson, and their measurement has been carried out by Miss Whitney. The observing program has been temporarily discontinued because the presence of large numbers of sunspots makes the interpretation of the results uncertain.

RELATION BETWEEN SOLAR AND TERRESTRIAL PHENOMENA

In collaboration with Dr. Oliver Wulf, of the U. S. Weather Bureau, Nicholson

has found evidence that typical recurrent geomagnetic activity is initiated when, owing to its rotation, relatively long-lived patches of bright flocculi on the sun come into view around its east limb. A mechanism for the production of this geomagnetic activity has been proposed involving the precipitation of circulatory instability in the upper atmosphere by the heating of absorbed ultraviolet radiation, the resulting irregular air motions in the ionosphere generating, by dynamo action, the electric currents whose fields constitute the geomagnetic activity. The dying-down of the activity after onset and while the patch of flocculi is still on the visible side of the sun follows from two causes: the decrease of electrical conductivity of the air incident to expansion, and the dying-away of the air motions themselves as the circulatory instability spends itself.

Series of periods of magnetic activity occurring at 27-day intervals do not appear to be distributed randomly in time, but occur in such a way that the onsets come preferentially near the times of high northerly or southerly lunar declinations. This phenomenon is interpreted as arising from the increased instability of air circulation in the upper atmosphere caused by the asymmetry of the air tide with respect to the earth's rotation at high lunar declination. This interpretation at the same time offers an unforced explanation of the well-known seasonal dependence of magnetic activity, which is pronounced near the equinoxes, since then the sun is changing the form of the atmospheric circulation most rapidly.

PLANETARY INVESTIGATIONS

A trail of a minor planet, discovered by W. Baade on a 3-hour photograph of the Crab nebula taken at the 100-inch telescope

on January 14, 1948, was so long that it seemed possible the object might be fairly close. A preliminary orbit, however, re-

vealed that it was distant 1.50 astronomical units. An orbit based upon an arc of 57 days was computed by Richardson together with an ephemeris for the next opposition, in July 1949. The elements and the ephemeris were sent to the Cincinnati Observatory, where the minor planet has been temporarily designated 1948 AA.

Comet 1925 II is remarkable for its low eccentricity (0.135) and its sudden changes in brightness of as much as six magni-

tudes. It has been suggested that these changes arise from exciting emission from the sun. Photographs of Comet 1925 II were taken by Richardson at the 60-inch telescope but failed to catch the object during a sudden change in brightness. A continuous study of this remarkable comet will be needed to understand the cause of its sudden changes. Occasional photographs add little to the work already done.

STELLAR INVESTIGATIONS

PHOTOELECTRIC PHOTOMETRY

Using the photoelectric photometer, whose development is described below under "Instrumentation," Pettit has made extensive measurements of stellar magnitudes. A large number of determinations of equivalent photographic magnitudes of stars in Selected Areas in the 30° zone have been made to implement the nebular program. Central stars in 13 areas and the field stars in SA 57 and SA 61 have been investigated.

Zeta Aurigae. Most of the available observing time from December to March was devoted to the eclipse of the B-type component of this star. Yellow, ultra-violet, and silver filters were used. Comparison stars were λ Aurigae and ρ Aurigae. It was found that the occultation phase showed an amplitude of 0.18 mag. in the yellow, 0.68 mag. in equivalent photographic light, and 2.02 mag. in the ultra-violet. The silver filter, because of light leakage, showed only 1.52 mag. increase during the occultation phase.

The star was tested 40 days after the occultation phase when the B-type component was shining through the outer envelope, about 0.9 diameter from the giant K5 star. These measures show that the absorption of light by the outer envelope near the limb of the K5 star was inap-

preciable in yellow (visual) light, but amounted to 0.07 mag. in equivalent photographic light and 0.12 mag. in the ultra-violet. The revised elements of the occultation phase are:

Epoch of minimum	JD 2432533.65
Period	.. 972.07d
Duration of eclipse	.. 39.50d
Duration of totality	36.80d

The visual magnitudes of the components reduced from the yellow-filter measures are 4.12 for the K5 star and 5.98 for the B-type companion.

VISUAL MAGNITUDES OF NOVAE

The visual magnitudes of Nova Puppis 1942 and T Coronae Borealis have been measured at intervals throughout the year. Nova Puppis is declining very slowly. Its present magnitude, 11.27, is only slightly lower than the average, 11.22, for last year. T Coronae Borealis also is declining slowly. Its present magnitude, 9.88, is still 0.3 mag. brighter than the level set by the K-type companion.

THEORY OF INTERIORS

An investigation of the effect of noise—the transfer of energy from the interior of a star by macroscopic mechanisms such as shock waves and turbulence, as distin-

guished from the steady energy flow due to heat conduction or radiation—in the convective region near the center of a star has been carried out by M. Schwarzschild

and Richardson. Integrations of the temperature, pressure, and mass of four selected stellar models have been completed and are being discussed by Schwarzschild.

STELLAR SPECTROSCOPY

RADIAL VELOCITIES

Excellent progress has been made by R. E. Wilson toward the completion of the five older Observatory radial-velocity programs mentioned in last year's report. About 200 plates are still needed to complete the observations of 140 stars, 120 of which are in the winter sky. During the report year 583 plates were taken, largely by Wilson, in connection with these programs and with a supplementary list of M-type stars selected to fill out the gap in the summer sky arising from the completion of the older program. In addition, about 480 spectrograms were obtained with the 60-inch telescope in connection with special investigations. The total of 1060 spectrograms taken with this instrument, compared with 800 taken during the preceding year, reflects in part an improvement in observing conditions and in part shorter average exposures.

Progress has also been made on the measurement of the plates. Whereas at this time last year there were on hand some 1700 plates in need of measurement (1300 unmeasured and 400 for remeasures), all but 50 of the plates taken this year have been measured, and the total unmeasured plates now number only 400. About 600 remeasures, however, are desirable and are under way.

Radial velocities of about 1500 stars have been completed. Those of 200 stars in the Taurus region were published by R. E. Wilson, and a manuscript containing the radial velocities and revised spectral classifications of 1300 others is being prepared

for publication by R. E. Wilson and A. H. Joy.

As was stated in last year's report, publication of a general catalogue of radial velocities is planned. As a first step in preparation, a card catalogue of radial velocities from all sources has been started and is being kept up-to-date. It is proposed to make January 1, 1950 the closure date for material which is to be included in this general catalogue. By that time it is hoped that all determinations of radial velocity which have not already been published will have been sent to us.

In 1944, S. A. Mitchell began at the 60-inch telescope a program of spectroscopic observation of certain dwarf stars which were being observed for trigonometric parallax at the McCormick Observatory. The observations have been completed, measures of radial velocity and estimates of spectral type have been made, and the material has been prepared for publication by Joy. Although the stars are limited to certain regions of the sky which could be observed in the summer and autumn, the results are a valuable addition to those obtained from 180 dwarf stars published in 1947.

STANDARDS FOR SPECTRAL TYPE

A tentative list of stars of various spectral types in the supergiant, giant, and main-sequence divisions has been prepared by Joy and distributed to observers of stellar spectra in the hope that the Henry Draper system of classification may thus be more specifically defined and be made more readily accessible.

VARIABLE STARS

Sanford has extended his observations of class N and Cepheid variables using the higher dispersion of the 32-inch camera of the coudé spectrograph. Particular attention was given to SV Vulpeculae, which is the Cepheid of the longest period (45 days) bright enough to be observed with this dispersion, and to W Virginis, the Cepheid which for various reasons is suspected of belonging to stellar population II.

Spectrograms of 30 variables of the RR Lyrae type have been obtained by Joy in addition to those previously published. Observations of certain stars of the RV Tauri, SS Cygni, and R Coronae types have been continued. The study of the spectra of variables in the globular clusters with periods greater than a day has been continued; 32 variables have been observed. The RR Lyrae, W Virginis, and RV Tauri types of variation are commonly found, but the Cepheid and the Mira types of variation, with few exceptions, are absent from the clusters. This distribution indicates that population II predominates in these systems.

Long-period variables. Data concerning numerous atomic absorption lines measured in a general investigation of the spectra of long-period variable stars of types Me and Se have recently been published by Merrill. The wave lengths, based on spectrograms having a dispersion of 10 Å/mm (20 Å/mm in the red), should have relatively few errors in excess of 0.02 Å, although blending and actual displacements cause many larger differences from the corresponding laboratory wave lengths. In spectral type Me the regions covered were $\lambda\lambda 3442-4575$; in type Se, $\lambda\lambda 3342-5064$ and $\lambda\lambda 5797-6832$.

Z Andromedae. In the rich shell spectrum of 1946, the large numbers of dark lines of ionized titanium and of ionized

chromium were noteworthy, and the displacements of the well defined dark lines in the Balmer series of hydrogen showed an interesting progression with quantum number. In 1947, no dark lines were present. Radial velocities derived from the bright lines suggest damped oscillations with a period of 700-750 days. Fluctuations shown by forbidden lines seem to be out of phase with respect to those shown by permitted lines. Various observed effects may arise from differences in the motions of atoms at various levels of the extensive stellar atmosphere. Wave lengths measured for a few forbidden lines of highly ionized iron atoms, [Fe v] and [Fe vii], may be more accurate than the calculated values.

Zeta Aurigae. O. C. Wilson has completed the analysis of the spectrograms of the 1939-1940 eclipse of Zeta Aurigae. During the eclipse of December 1947 and January 1948, an excellent series of spectrograms was obtained covering both ingress and egress, in spite of much poor seeing and some clouds. A superficial inspection shows that the behavior at ingress was quite different from that at egress. The ingress of 1947-1948 was very similar to the ingress of the 1939-1940 eclipse, but the egress differed substantially from that of the earlier eclipse. At least as late as 8 days after the fourth contact all the lines were very much weaker than at the corresponding phases of ingress. Pending a more thorough analysis of the results, one is led to the tentative conclusion that there was a considerable difference in the amount of matter at corresponding heights on the two sides of the K-type star.

STARS WITH EMISSION LINES

Early types. In the program of Merrill, Burwell, Miller, and Mowbray for the discovery and observation of bright-line stars

of early type, the emphasis during the past year has been on (1) listing the bright *Ha* objects visible on our objective-prism photographs and (2) obtaining slit spectrograms for as many as possible of the new ones. On July 1, 1948, the lists included 250 stars for which no slit spectrograms had yet been obtained.

Stars with emission calcium. A list of 250 stars, both giants and dwarfs, showing emission H and K lines of calcium is being prepared by Joy and R. E. Wilson. Since the spectrum at $\lambda 3950$ is, in general, poorly exposed on the spectrograms, the list is far from complete, but will serve to show that calcium emission is not an uncommon phenomenon in stars of various classes.

Wolf-Rayet stars. O. C. Wilson has nearly completed his survey of Wolf-Rayet stars available with the 60-inch. This program has two objectives: (a) to obtain a minimum of 5 spectrograms for each W star not adequately observed elsewhere, in order to find new binaries, if possible, and thus to cover a large enough sample of the W stars to give a good idea of the percentage of binaries among them; and (b) to utilize all the plates so obtained to investigate the curious displacements of some of the emission bands previously observed in some of the W binaries by both Hiltner and Wilson. As to (a) it may be stated that very few new binaries have been found. Two or three of the stars are of probable, though not yet certain, binary character; none has a very large amplitude. These stars are undergoing further investigation. A number of stars characterized by bright 4686 *He II* and 4634-4640 *N III* have been included in the observing list.

The binaries under investigation include HD 190918, HD 193793, HD 165688, and BD + 40° 4220. HD 50896 should be included although it is not certainly a binary, even though a good deal of variation has

been found in its spectrum. Although none of these investigations is complete, the period of HD 190918 appears to be about 85 days, which, if confirmed, will be the longest period yet found for a W binary.

Novae. Zwicky, with the assistance of Johnson and A. G. Wilson, is engaged in a preliminary objective-prism survey of the Milky Way out to galactic latitudes $\pm 25^\circ$, using the 18-inch Schmidt camera on Palomar Mountain. The primary object of this survey is to lay the basis for study of the galactic aspects of the general problem of the distribution, frequency, and classification of novae. After completion of the preliminary survey, it is hoped to establish a control period of about four months, with the object of detecting all galactic novae brighter than tenth magnitude occurring in this time interval. Particular study is being made of the effects of selectivity on discovery of novae.

The preliminary survey is threefold: (1) a prism survey in the blue ($\lambda 3000$ to $\lambda 5000$) to about the eleventh magnitude; (2) a prism survey in the red ($\lambda 5000$ to $\lambda 6900$) to about magnitude 12.5; (3) a direct survey to about the sixteenth magnitude for reference purposes. By-products of the survey have included the discovery of new emission nebulae and the chance photography of several meteor spectra.

Merrill, Burwell, and Miller made a short investigation of the spectrum of Nova Sagittarii 1947, whose discovery was mentioned in last year's report. The outward velocity of the expanding atmosphere was found to be 1050 km/sec, and the velocity of the star as a whole (freed from solar motion) was +190 km/sec. The nova lies in a direction nearly at right angles to the galactic motion of the sun; hence the rapid recession must be due to its own motion.

Sanford has continued the series of observations of the recurrent nova T Coronae Borealis. The spectrum and magnitude have changed little during the past year. The emission lines of H , He II, N III, O III, Ne III, and Ne V are readily seen overlying a class-M spectrum that is well shown in the blue, green, yellow, and red regions of the spectrum. Radial velocities from the absorption lines of the class-M spectrum continue to conform to a variation in a period of somewhat more than 200 days.

Spectra of individual stars. Interesting changes were found by Merrill in the spectrum of HD 236031, a bright-line star discovered at Mount Wilson in 1944. A detailed study of the variable components of the Balmer series of hydrogen lines in the spectrum of HD 45910 is nearly ready for publication. A long series of spectrograms of 48 Librae indicates that an oscillation in the star's atmosphere which began in 1934 has since been subject to strong damping. Additional spectrograms of a number of other peculiar stars have been obtained during the year. In a few shell stars, progressive changes in velocity from line to line along the Balmer series may provide a means of studying accelerated motions in the stellar atmospheres.

GENERAL MAGNETIC FIELD OF EARLY-TYPE STARS

Observation of stellar magnetic fields has been continued by H. W. Babcock with emphasis on the "peculiar" stars of type A, and particularly on the spectrum variables that form a subgroup of this type. The best example of a spectrum variable accessible to the 100-inch is HD 125248 (BD-18°3789). This star has been found to have an alternating field that varies between the approximate limits +7800 and -6500 gauss, with a regular period

of 9.295 days. The magnetic changes are synchronous with the pronounced changes in line intensity previously observed by Morgan and Deutsch. A detailed study of the magnetic variations and other spectral peculiarities of this star is in progress.

Many of the abnormal line intensities and their variations in the peculiar stars of type A can be explained at least qualitatively in terms of Zeeman broadening of strongly saturated absorption lines. Some lines of Eu II, normally of width 0.023 Å, may attain an actual width of 0.35 Å in a magnetic field of 8 kilogauss, and may have their total absorption increased by a factor approaching 8. Considerable quantitative work on this effect should be profitable. Since rotational broadening does not alter the equivalent width of a line, it is possible at least in principle to study the magnetic fields of stars by analyzing the magnetic intensification of absorption lines, even for cases in which rotational broadening does not permit the use of the circular analyzer. It has been recognized for some years that the ionization theory alone cannot account for the abnormal line intensities in the peculiar A stars, and it now seems that magnetic intensification should go far toward resolving these difficulties without recourse to vastly abnormal abundances of the elements.

Magnetic effects have been measured or strongly suspected in practically all the sharp-line spectrum variables and peculiar stars of type A that have been examined with the circular analyzer, and it may be inferred that all the spectrum variables have varying magnetic fields. In some of the peculiar stars, constant abnormal line intensities are found, together with a magnetic field that is constant within the observational error. An example is γ Equulei, for which six plates obtained in 1946 and 1947 give $H_p = 1900 \pm 250$ gauss. In this

star the magnetization and rotation are found to be antiparallel on the assumption that the star rotates on its axis in the direction in which its companion revolves in its orbit.

About 25 stars are being followed when opportunity permits, and test plates of other suspected objects are taken when possible. Even though all these spectra cannot be measured in detail as rapidly as they are taken, they are quite valuable for reference. Eventually it may be possible to find relations between such parameters as field strength, period of variation, specific spectral peculiarities, rate of axial rotation, and others.

The observed variations in the general magnetic field of HD 125248 and of some other spectrum variable stars constitute evidence for specific sources of cosmic rays according to either the Cygnatron process proposed by Swann or an extension of the model of Davis. The observations of Forbush, of the Department of Terrestrial Magnetism, indicate that the Cygnatron action operates to accelerate electrons in sunspots; if this is so, similar action may be of importance in many stars, even those with a relatively weak but fluctuating magnetic field.

THE RELATIVE ABUNDANCE OF THE CARBON ISOTOPES

Sanford and O. C. Wilson have obtained spectrograms of some of the brighter class-R stars in which bands of both $C^{12}C^{12}$ and $C^{12}C^{13}$ occur. The relative intensities of these two types of bands and data for the relative transition probabilities for vibrational heads of the $C^{12}C^{12}$ bands obtained by King in the laboratory offer a possible new approach for deriving the abundance of C^{13} relative to C^{12} in this class of stars.

An attempt was made by O. C. Wilson

to set a lower limit to the $C^{12}C^{13}$ ratio in interstellar space by utilizing the excellent 9-foot coude spectrograms of ζ Ophiuchi taken by Adams. The result obtained was $C^{12}C^{13} \geq 5$. Necessarily this is somewhat in the nature of an estimate and hence is not precise. It is hoped that eventually the 200-inch telescope may considerably improve and extend knowledge on this very important point.

INTERSTELLAR LINES

The results of a study of interstellar lines in the spectra of 300 stars of types O-B8 are being prepared for publication by Adams. The photographs were obtained with the coude spectrograph of the 100-inch telescope, about three-fourths of the stars being observed on a linear scale of 2.9 Å/mm. The study includes examination and measurement of the structure of the H and K lines, investigation of the presence or absence of other atomic or molecular lines, estimates of intensity, and comprehensive measurements of radial velocity. The results are being applied to identifications of gaseous interstellar clouds through their motions, to the question of the place of origin of the molecular lines, and to a discussion of the galactic rotation as determined from the interstellar clouds.

MISCELLANEOUS

The A-type star HD 161817 was found by Albitzky at the Simeis Observatory to have the extremely high velocity -360 km/sec. Recent observations by Merrill have confirmed this result and have shown that the spectrum closely resembles that of a sharp-line main-sequence star of type A4, absolute magnitude +1.

In a brief descriptive survey, Merrill has listed twenty types of stars whose spectra exhibit recurrent changes.

The radial-velocity variation of the very long-period spectroscopic binary Boss 1074, which Sanford had previously concluded to have a period of 6270 days, has now been found to require over 10,000 days to com-

plete. Thus Boss 1074 has, if not the longest, one of the longest periods of velocity variation established wholly by radial velocities well distributed over an entire cycle.

GALACTIC NEBULAE AND STAR CLOUDS

PLANETARY NEBULAE

Minkowski's survey of planetary nebulae is now virtually completed. During the past year, 57 new planetaries have been identified. The total number of planetaries found up to date on the 10-inch objective-prism plates is 168. For a few selected fields, objective-prism exposures obtained with the 18-inch Schmidt camera on Palomar Mountain by J. C. Duncan and by A. G. Wilson have become available and are being used to determine the degree of completeness of the 10-inch material. The investigation of objects found on the Schmidt-camera films has led to the discovery of 12 additional planetaries. With the addition of these 180 new objects, the total number of planetaries known is now 335.

A search for planetary nebulae in globular clusters. Since it has become clear from Minkowski's investigations that the planetary nebulae belong to population II and since the globular cluster Messier 15 contains a planetary nebula, Baade has searched for planetaries in eight of the nearer globular clusters, using the same technique with which he finds emission nebulae in the Andromeda nebula. No planetaries were found in this preliminary survey, a fact which suggests that these objects are rare in globular clusters. Since even the richest globular clusters are relatively small samples of population II, the result was almost to be expected.

Survey of spectra with high dispersion. The first stage of the high-dispersion sur-

vey of the spectra of the brighter planetaries by O. C. Wilson is also approaching completion although considerable observing remains to be done. Perhaps the most interesting object yet worked on is NGC 2392, where the processes of ejection can be traced from the reversing layer of the nucleus through the region where the nuclear emission bands are formed, out into the nebula. The measures indicate that the stellar reversing layer is highly stratified in the sense that lines of different excitations are formed at different depths, and is characterized by an outward velocity which is a function of depth. These results render very doubtful the value of studies of the abundances of elements in the nuclei based on the assumption of static atmospheres.

Results on the nebular portion of the spectrum of NGC 2392 are perhaps even more spectacular. The lines of $[O\ II]$, $[O\ III]$, H , and $[Ne\ III]$ are double, with a separation between the components of about 100 km/sec. The lines of $[Ne\ V]$, on the other hand, are single and correspond in displacement to the means of the two components of the double lines. Hence the atoms producing the lines of $[Ne\ III]$ and those producing the lines of $[Ne\ V]$ are clearly two separate and distinct groups, and it is meaningless to compare the intensities of $[Ne\ III]$ and $[Ne\ V]$ in studies to determine physical conditions in the nebula or abundances of elements. In spite of this obvious difference of behavior of $[Ne\ III]$ and $[Ne\ V]$, the slit-

less images given by the two lines are identical in size and shape within the limit of errors of measurement.

The same behavior of $[Ne\ v]$ has been found in NGC 6741. Existing plates suggest that the lines of $[Ne\ v]$ have the same structure in three or four other nebulae, although longer exposures will be required to confirm this.

VARIABLES IN THE NUCLEAR REGION OF THE GALACTIC SYSTEM

Baade has continued the observation of the variables in the field around the globu-

lar cluster NGC 6522 ($\lambda = 328^\circ$, $\beta = -4^\circ$). Two extended runs of observations, to be carried out in July and August 1948, should provide the necessary data for the numerous cluster-type variables, but the determinations of the periods of the long-period variables will require scattered observations during the next few years, since it cannot be decided from the present material whether the periods are of the order of 360 days or 180 days. A start has been made on transferring the photographic scale of Selected Area 68 to the region.

EXTRAGALACTIC NEBULAE

PHOTOELECTRIC MAGNITUDES AND COLORS

The photometry of the extragalactic nebulae for which radial velocities have been obtained was begun in August 1947 by Pettit, and the equivalent photographic magnitudes on the International scale for 109 of these objects have been reduced. For

Eight nebulae for which the same apertures were used are common to this list and that of Stebbins and Whitford; the average residual without regard to sign is 0.06 magnitude. The reduction of colors is not yet completed.

THE STRUCTURAL PATTERN

Hubble has made considerable progress in preparing for publication a study of the structural pattern in the thousand brightest extragalactic nebulae, and in selecting typical examples for reproduction. Observing time has been used largely for replacement of inferior plates and for attempts to get additional information with ultraviolet and red exposures.

Catalogue m _{ph}	No. nebulae	(O - C) mag.
9.....	2	-0.32
10.....	14	-0.43
11.....	31	-0.17
12.....	32	-0.14
13.....	13	+0.26
14.....	17	+0.22

most of these, estimates were available from the Shapley-Ames catalogue, but for systems fainter than magnitude 13.3, estimates from the plates taken at Mount Wilson were available. The mean differences between the photoelectric and catalogue values (O-C) are shown in the accompanying table. Some deviations of as much as a magnitude have been found between the present measures and the catalogue values, but the photoelectric values have been verified by further measurements.

SUPERNOVA IN NGC 4699

During this investigation a supernova was found in NGC 4699, an intermediate-type spiral in the south lobe of the Virgo Cluster. The nova, 40 seconds of arc north of the nucleus, was followed from March 5 to April 4, 1948, while the magnitude dropped from 17.0 to 18.2. Since the modulus of the cluster is $m - M = 26.8$, the nova evidently was a few months past maximum when discovered.

SURVEY OF THE ANDROMEDA NEBULA

In the fall of 1947, Baade extended his survey of the Andromeda nebula with ultraviolet, blue, red, and near infrared exposures to the north preceding outer region, up to $2^{\circ}.5$ from the nucleus. This leaves for the coming season the heretofore unexplored outer regions in the neighborhood of the minor axis. Besides yielding a large number of emission nebulae and clusters, the survey has revealed a most interesting feature of the Andromeda nebula. Population II, which at first seemed to be restricted to the central region of the nebula where it predominates, actually extends through the whole system. Beyond the "amorphous" central lens it becomes at first lost in the strong innermost spiral arms, but farther out it emerges very clearly again and again, with decreasing intensity, in the clear spaces between subsequent spiral arms. In fact, it is easily traced beyond the outermost faint spiral arms. These observations suggest that as far as mass is concerned, the main substance of M 31 is provided by population II, the spiral structure (population I) embedded in it playing the role of an incidental feature. It also seems certain now that the faint outer extensions of M 31 which Stebbins and Whitford first detected some 15 years ago are identical with the outermost fringe of population II.

NUCLEAR EMISSIONS OF EXTRAGALACTIC NEBULAE

To broaden the basis for a discussion of the emission spectra appearing in the nuclei of some spirals, Minkowski obtained spectra of some extragalactic nebulae with medium dispersion in the photographic region and with a low-dispersion grating spectrograph in the visual region. Emission lines of H , $[N\text{ II}]$, and $[O\text{ III}]$ with a total width corresponding to a velocity of

700 km/sec have been found in the nucleus of NGC 3031 (Sb). Narrow faint H emissions are present in the nucleus of NGC 4535 (Sc), this nucleus having a high luminosity similar to those of the nebulae investigated by C. Seyfert several years earlier, which show strong emission lines of considerable width. The new results seem to indicate that the width of the nuclear emissions is not correlated with the luminosity of the nucleus.

SPECTRA OF EXTRAGALACTIC NEBULAE

Humason has continued the preparation of 500 nebular velocities for publication. At present all velocities obtained at Mount Wilson with low dispersion (500 Å/mm at $H\gamma$) are being revised with the use of improved wave lengths derived from an extensive series of measures on spectra of NGC 221. New wave lengths have been computed for the following absorption features, old values being shown in parentheses: $\lambda\lambda 3885.37$, (3889.05); 4304.37 , (4303.14); 4384.96 , (4383.56). Observational time has been entirely devoted to reobserving nebulae for which only poor spectrograms were available. Improved spectra of 46 such objects have been obtained during the year.

RADIAL VELOCITIES AND SPECTRAL TYPES OF GLOBULAR CLUSTERS IN MESSIER 31

To investigate the spectral types and state of motion in the system of globular clusters associated with M 31, Humason started observations of a selected list of 17 clusters brighter than magnitude 16.0. Velocities of 10 clusters have so far been measured. The dispersion used is 146 Å/mm at $\lambda 3900$, and the mean error of the results for the 10 objects so far observed is ± 18 km/sec. The mean number of lines measured on an individual plate is 11, the large majority being in the region of H and K or farther in the violet.

LABORATORY INVESTIGATIONS

TRANSITION PROBABILITIES

The measurement of the relative *gf*-values for 134 lines in 37 multiplets of the spectrum of neutral nickel (*Ni* I) has been completed by R. B. King. The data were derived from electric-furnace absorption spectra; hence the observations were limited to lines arising from the three lowest terms in the atom with an over-all separation of less than 0.5 volt. The list includes, however, most of the lines of astrophysical importance in the region $\lambda\lambda 3012\text{--}3912$. The measured *gf*-values reveal wide departures from normal multiplet-line strengths on the basis of LS coupling.

The relative transition probabilities of 20 vibrational bands belonging to five sequences in the Swan system of the carbon molecule C_2 have been obtained from measurements of peak intensities of the band heads in furnace emission spectra. The results are in fairly good agreement with theoretical values for a symmetrical diatomic molecule. Work on the furnace spectra of C_2 is being continued in an effort to determine absolute transition probabilities of the Swan bands and to establish the ground state of C_2 , which is open to question at the present time.

Mr. William C. Carter, of the California Institute of Technology, has continued the direct measurement of line intensities in electric-furnace emission spectra of *Fe* I with the aid of a photomultiplier tube. Preliminary results indicate that the furnace is a sufficiently stable source to make this method practicable, and that the photomultiplier tube is sufficiently sensitive to detect and measure the intensities of many more lines from combinations between higher energy levels than could be obtained in absorption spectra.

The measurement of absolute transition

probabilities by the method of total absorption of the weak intersystem ($^1S\text{--}^3P$) lines from the ground states of *Mg*, *Ca*, and like atoms is being undertaken in the laboratory by Mr. Frank Estabrook, of the California Institute of Technology. The transition probabilities of these lines are of great importance in the problem of the stellar abundance of these elements.

THE SPECTRUM OF DYSPROSIUM

A. S. King has continued his study of the spectrum of dysprosium. Intensity estimates have been made and impurity lines eliminated in the region from $\lambda 4600$ to $\lambda 9400$.

INSTRUMENTATION

The mechanical parts of a new polarizing compensator designed by H. W. Babcock have been built in the shop; when this instrument has been completed and calibrated, it may permit more effective observations with the circular analyzer of stars in the northern part of the sky.

The machine shop has completed mechanical work on the new direct-intensity microphotometer, except for a few minor details, and the construction and wiring of the electrical units, which include some 36 tubes, have likewise been essentially completed by the electronics shop. In general, the performance of the machine seems to meet the specifications laid down when the design was begun, and it appears that no major changes will be required. Acceptable tracings of coudé spectrograms (2.9 Å/mm) having numerous sharp lines can be made at a scanning rate of about 3 mm per minute, and in many cases considerably higher speeds can be used. Final adjustments are yet to be made on some parts of the electronic circuits, and

one or two of the mechanical parts may require some further attention. The machine should be ready for routine operation in a few months. The associated calibration spectrograph may, however, require further checking before its linearity can be relied upon.

The automatic guider has continued in regular use with little attention; various ideas for simplifying the adjustments and improving the design are being considered.

An improved experimental model of the integrating exposure meter for the coude spectrograph has been tested, but time has not permitted the building of a reliable model for regular use.

A photoelectric photometer using a 1-P21 multiplier tube was constructed by Pettit for the photometry of extragalactic nebulae at the telescope. The salient features of this photometer are (1) refrigeration, (2) a quartz lens system to transfer the field light to the cathode, (3) a single-stage amplifier, (4) field illumination at the Cassegrain aperture, (5) 19-second and 6-second galvanometers to measure the signal.

Laboratory tests of the properties of the photometer show that refrigeration with dry ice increases the useful range of the instrument 4 magnitudes when the exter-

nal temperature is 0° C; at the same time there is a drop in specific sensitivity. The sensitivity of the instrument is such that anything that can be seen in the telescope can be measured. With the 60-inch telescope stars as faint as magnitude 16.5 are measurable. The usable range, however, is considerably affected by high humidity.

The uniformity of response of the photometer to light was tested with a polarimeter over the range 5 to 17 magnitudes equivalent deflection with the 60-inch telescope and found to be satisfactory. The response of the light transmitted by the various apertures used on the nebular programs was also tested. The photometer is provided with filters for the yellow, blue, and ultraviolet. Tests made on the north polar sequence show that the free-minus-yellow deflections on a logarithmic scale give photographic magnitudes over the range tested, 9 to 16 magnitudes. Color index on the International scale can be obtained from either the ratio yellow to free-minus-yellow, or the ratio yellow to blue.

Two gratings, each over 7 inches wide, have been ruled by H. D. Babcock, assisted by Prall. These show essentially their theoretical resolving power in the first six orders.

THE LIBRARIES

The year 1947-1948 has been one of growth and activity largely brought about by the reorganization connected with the beginning of joint operation of the Mount Wilson and Palomar Observatories. First to be mentioned is the organization of a new astronomical library in the Robinson Laboratory of Astrophysics on the campus of the California Institute of Technology. At the end of the first year this library contains about 4000 volumes, and is already

a useful working collection, although still undeveloped in some sections.

The nucleus of the collection was formed by hundreds of volumes from the libraries of Dr. Hale and Dr. van Maanen. These volumes, now marked with appropriate bookplates to indicate their source, include many separate books, in addition to long runs of periodicals and observatory publications which would be very expensive and difficult to acquire by purchase. Most of

them are already bound. Another part of the collection consists of volumes transferred from the General Library and from the Physics Library of the California Institute. Numerous other volumes, particularly the more recent books, have been purchased. This new library is housed in a room which was especially designed for this purpose and is very beautiful in its architectural details. The collection is planned to meet the ordinary needs of Observatory staff members with offices on the Institute campus and of students in the astronomy department, and is not intended to duplicate the much larger library at the Mount Wilson Observatory offices on Santa Barbara Street, which will remain the principal reference library.

In addition to the two main libraries, both located in Pasadena, small collections are maintained on Mount Wilson and Palomar Mountain for the use of observers.

During the year 1947-1948 the library on Santa Barbara Street has acquired 332 volumes, 156 by binding, 52 by purchase, and 124 by gift, making a total of 16,330 accessioned volumes. The gifts are almost all volumes which have been accessioned and catalogued during the current year from the Hale and van Maanen libraries. The bequests of both these libraries have been recorded in recent annual reports. As has already been noted, duplicates from these libraries form the nucleus of the new library in the Robinson Laboratory of Astrophysics.

The Hale library contained many rare and old books, association books, and

volumes on biography and the history and philosophy of science and kindred subjects. These are being incorporated in the Santa Barbara Street library. Unfortunately, there has been no suitable place to use or display these rare books from the Hale library or the rare books already owned by the library which were purchased largely because of Dr. Hale's interest in the field of the history of astronomy while he was Director of the Observatory.

In the spring of 1948, however, alterations were made to provide what is known as the "Hale Room." This room, remodeled to open only into the librarian's office, has book shelves filling all available wall space almost to the ceiling. Instead of a door, the room has a wrought-iron grille with its name, "Hale Room," and the Egyptian sun symbols worked into the design. For further protection of the older books the shelves on one wall have grille doors. The dark finish of the woodwork, the black grilles, and the brass fittings combine to give a rich and pleasing background for the fine old books which were given by Dr. Hale or purchased by the Observatory. From Dr. Hale came the rarest items, such as the 1600 edition of Gilbert's *De magnetice*; first editions of the works of Tycho Brahe, Kepler, and Galileo (*Sidereus nuncius*, 1610; *Macchie solari*, 1613; *Dialogo dei due sistemi del mondo*, 1632; etc.); Scheiner's *Rosa ursina*; a fine run of other astronomical books from the sixteenth through the eighteenth century; and many important modern works.

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GEOPHYSICAL LABORATORY

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Studies of the earth have as their objective the determination of the nature of the processes whereby the whole earth and the materials of which it is composed have come into being and have acquired the forms, the disposition, and the mutual relations in which we find them. If we may be permitted to borrow terms from our biological colleagues and to expand their meanings somewhat, it might be said that earth processes are studied *in vivo*, *in vitro*, and *post mortem*.

The first approach, observation *in vivo*, is the most direct but, in the nature of things, is available only on special occasions and for some processes. The transportation of sediment by a river and its deposition as a delta at the river's mouth is a process that can be so examined; and it is easy to estimate what net result may be expected when this action goes on for some millions of years. From such a relatively unexciting study there are all gradations of dramatic interest up to the study of a volcano in actual eruption, which may include examination of the physical and chemical characters of the materials erupted—rocks and gases—and the measurement of temperatures, of accompanying earth movements, and of any other characters that lend themselves to measurement.

Apart from their intrinsic interest, penetrating studies of processes which can be examined directly are of immense value in facilitating the interpretation of other processes which, because they have long since been completed, can be studied only *post mortem*. Actually, the latter kind of examination is not a study of a process, but rather observation of its results or end

products, from the characters of which it is hoped to determine the nature of the process. Thus, when we find in ancient terranes masses of rock which have crystalline characters, flow structures, gas vesicles, and other features analogous to those of rocks which are poured out today from volcanoes such as Paricutin, we may feel sure that these particular ancient rocks were formed in a similar manner.

There are, however, some inaccessible highly important geologic processes that we may not hope to observe while they are occurring, such as those that take place in the depths of the earth or even at moderate distances below the surface. The products of these processes are sometimes brought to the light of day and come under our observation; and again we attempt to deduce the nature of the processes from the character of the product, but for this examination there are no strictly pertinent observations of current phenomena to serve as a guide, with the result that alternative interpretations may be possible. It is here that studies *in vitro* come into play. By subjecting materials to a wide range of conditions in the laboratory and observing their behavior, we may be enabled to decide between alternative views as to the manner in which they have acquired certain characteristics.

The experimental studies are, in actual practice, carried beyond the simple initiative stage implied in the foregoing remarks. The behavior of a pertinent material is observed under carefully controlled and measured conditions in order that the laws of its behavior may be discovered and that broad generalizations

may thus be laid down. Such laws can then be used to predict its behavior under wide-ranging conditions, such, for example, as those prevailing in the depths of the earth.

The work of the Geophysical Laboratory has been and continues to be concerned principally with experimentation, though examination of living processes (e.g., volcanism) has a prominent place, and studies of the end results of ancient phenomena are far from neglected. Indeed, the principal studies of the earth carried out by field geologists must fall largely in the last category, and it is generally true that through field investigations the earth problems are first visualized and some of them come to be placed on the list of problems requiring laboratory investigation. For this reason, it is the fixed policy of the Geophysical Laboratory to keep in close touch with the current work of field geologists and to participate in it in some measure. The individual who is primarily a laboratory investigator may thus acquire an insight into the methods whereby the field geologist grasps his particular problems, and he may also be able to sharpen the tools of the field geologist by adapting experimental techniques to the measurement of quantities in the field. Moreover, after a field trip the laboratory man will return to his laboratory and conduct his investigations there, guided by a clear picture of the problems posed by the field study.

The supplementing of field studies by laboratory investigation is, in some instances, simply a closer determination of the properties and behavior of materials brought from the field to the site of facilities for making these determinations. Though conducted in a laboratory, such studies are not to be considered mere duplication of geologic processes, but

actually are refined *post-mortem* investigations. A significant proportion of the work of the Laboratory is of this kind, a sharpening of field observations and conclusions. Thus, granite specimens collected in New England, in the course of the determination of pertinent field relations, are subjected to examination in thin section under the polarizing microscope, and the exact nature and proportions of their constituent minerals are determined. The information thus afforded is subjected to statistical analysis in order to test alternative concepts of the origin of granite (Chayes).

Volcanic products of various kinds (rocks, gases, sublimates) collected during an *in vivo* study (Zies) are brought to this Laboratory for exact determination of their chemical and mineralogical characters (Zies, Merwin), in order that the information gained may be applied to the interpretation of other occurrences where there is no opportunity to observe and measure the conditions of formation of like materials.

Rocks of the peridotite clan have been studied with special reference to their radioactive content, in order to test certain alternative theories of the origin of such rocks (Davis). Quartz crystals from different rocks known to show a wide range of temperature of formation have been subjected to thermal analysis to test whether the inversion temperature from low to high quartz is related to the temperature of formation of the quartz (Tuttle).

In addition, the Laboratory has in a sense been taken into the field. Apparatus for measuring the variation of intensity of the earth's gravitational field has been set up at various points in the vicinity of Washington (England); and, in co-operation with the Department of Terrestrial Magnetism and the U. S. Navy, determinations have been made of the travel times of shock waves initiated by setting off charges of

high explosives, with the object of determining the velocity of travel in subsurface layers of the earth, from which it is hoped to ascertain the nature of the layers (Greig, England, Adams).

The studies with which the Laboratory is identified in scientific circles and which form the core of the Laboratory's program are those in which earth substances are subjected to various controlled conditions and the resultant effects are measured—true *in vitro* studies. In nature the substances are subjected to a variety of temperatures, pressures, and chemical environments, and they respond to these in a variety of ways, a record of which may be left in the material itself. It is the province of investigations of this type to subject these materials to a range of temperatures, pressures, and chemical conditions in the laboratory and to observe their response and the character of the record left in the material. In this manner it is hoped that the record of the natural materials may be interpreted and their conditions of formation deciphered. In the past work of the Laboratory the principal variable of which the effects were studied was temperature. Thus the temperatures of melting and inversion of a great many rock-forming oxides and of their compounds and mixtures of compounds have been determined, and work of that kind has been continued

(Schairer, Roedder). Just now greater emphasis is being placed on investigation of the effects of pressure and of pressure and temperature combined (Adams, Yoder) and especially of high pressures of water (vapor), which is known to be an important constituent of all rocks and even of the melts from which the primary igneous rocks form.

This problem of the behavior of rock-forming minerals with water at high temperatures and pressures has been under investigation for some time, the earlier studies concentrating on silicates of the alkalis (Morey) (Burlew). Studies are now being extended to other materials with the aid of a new type of apparatus, and results of great significance to problems of the origin of certain rocks have been obtained in investigations of the silicates of magnesia with water (Bowen, Tuttle).

This brief general picture of current work, its setting and its aims, is rounded out by studies that seek to establish the values of fundamental thermodynamic quantities for earth substances, such as heats of formation, heats of melting and inversion, and entropies, with the aid of which it is hoped to enable more complete prediction of the behavior of earth materials brought together under natural conditions of wide variety (Kracek, Sahama, Neuvonen).

SILICATE MIXTURES IN THE PRESENCE OF WATER UNDER PRESSURE

During the past year a number of definite conclusions have been reached in the course of the investigation of the stability relations of "low-temperature" minerals. These are minerals that from the conditions of their occurrence in nature appear to have formed, not at the elevated temperatures (1000° C or more) associated with the molten lava from active volcanoes,

but rather at significantly lower temperatures and in the presence of water and other volatile materials. Several lines of experimentation have already produced conclusive results. These procedures are being continued actively, and new techniques are being developed for pursuing further the studies of the genesis of the so-called "low-temperature" minerals.

THE SYSTEM MAGNESIA—SILICA—WATER

Investigation of the system $\text{MgO—SiO}_2\text{—H}_2\text{O}$, preliminary results of which were reported a year ago, has now been carried

pressure. One method for presenting the relationships is to draw a series of curves of univariant equilibrium as shown in figure 1. Each of the curves is of funda-

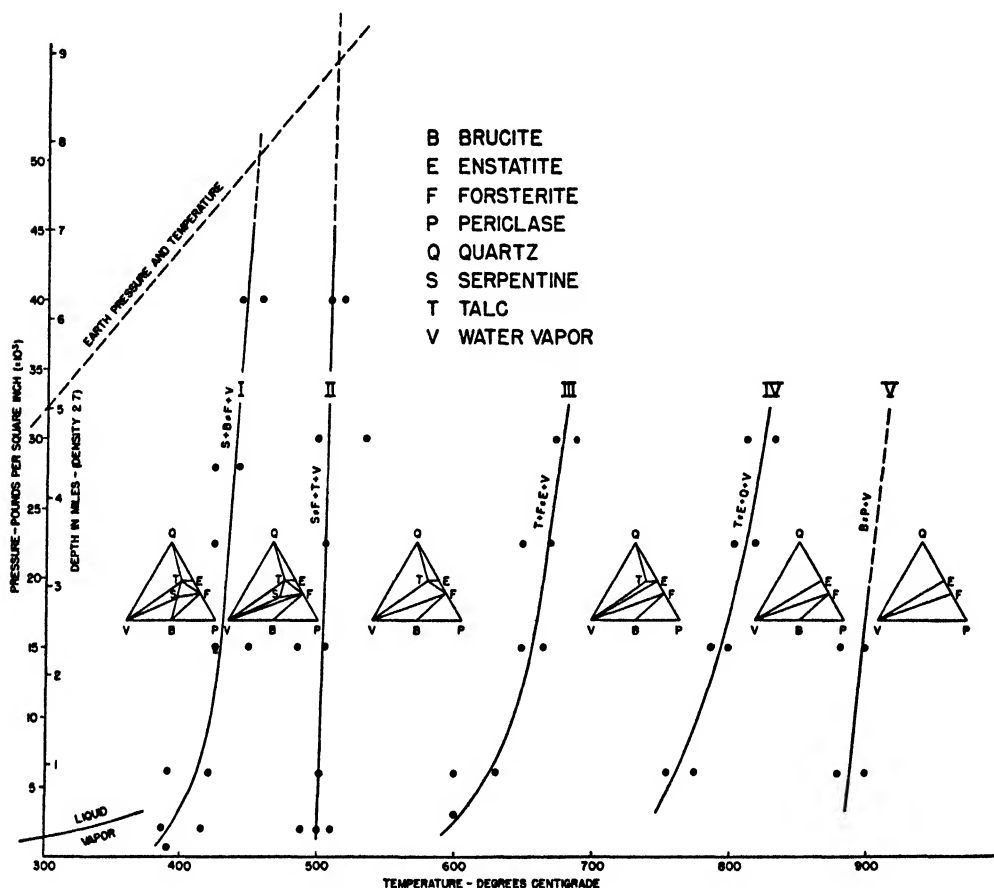


FIG. 1. Pressure-temperature curves (I-V) of univariant equilibrium in the system $\text{MgO—SiO}_2\text{—H}_2\text{O}$. Equation on each curve indicates the reaction to which the curve refers. Triangular diagram on each divariant region between curves indicates, for all compositions, the stable phase assemblages under the range of pressure and temperature conditions represented by the region. Each heavy dot indicates the temperature and pressure of a critical experiment which fixes the position of a curve. Lower left: vapor pressure curve of water ending at the critical temperature and pressure. Upper left: curve of normal earth pressure and temperature assuming a temperature gradient of 55°C/mi and a rock density of 2.7.

to the point where the results are ready for publication (Bowen, Tuttle). The range of stability of each of the crystalline phases has been determined under widely varying conditions of temperature and

mental significance in connection with the conditions of formation of the corresponding natural mineral assemblages in rocks. A few of the salient features shown by the diagram are indicated here.

Upper limit of stability of serpentine. Serpentine ($3\text{MgO} \cdot 2\text{SiO}_2 \cdot 2\text{H}_2\text{O}$) in its fibrous form, asbestos, is a valuable commodity, and a knowledge of the conditions of its formation may be of practical interest as well as of fundamental significance in earth science. It has been found that serpentine has an upper limit of stability at approximately 500°C , at which it decomposes reversibly into forsterite plus talc plus water vapor. The temperature of reaction is only slightly affected by pressure (curve II) between 2000 and 40,000 lb/in², corresponding approximately to depths in the earth of $\frac{1}{4}$ mile to 6 miles. This definite upper limit of temperature of formation of serpentine is useful in fixing the temperature of certain changes that have taken place in the rocks in which it occurs.

Serpentinization of olivine. One of the changes in rocks which have thus been determined as occurring below 500°C is the change of olivine to serpentine, but the actual relations are somewhat more complicated than might be inferred from this statement. The upper limit of stability of serpentine (500°C) is not also the lower limit of stability of magnesian olivine (forsterite) in the presence of water vapor. This lower limit of stability of forsterite, in respect to temperature and pressure, is given by curve I. Under the conditions pertaining to the region below this curve, forsterite is attacked by water vapor under pressure with production of serpentine and brucite. The temperatures now found are distinctly lower than have usually been assumed to prevail in the serpentinization of forsteritic olivine. The effect of the presence of iron in the olivine has been investigated by determining, with natural olivine, the temperature of the change represented by curve I, and it has been found that a natural olivine with 7.26 per cent FeO is changed to serpentine only at a

temperature some 60° lower than is pure forsteritic olivine. Plainly, serpentinization of natural olivine takes place at quite low temperatures.

Failure to obtain a serpentine melt. The possibility of obtaining a molten liquid of the composition of serpentine has been further investigated, because the existence of such a liquid magma at moderate temperatures has been assumed by some investigators. It had already been found that at 800°C and 30,000 lb/in² and also at 900°C and 15,000 lb/in² no liquid was formed in a mixture of the composition of serpentine. The product consisted only of forsterite and enstatite crystals. In order to check the possibility that the presence of iron might induce formation of liquid at moderate temperatures, a natural olivine containing 7.26 per cent FeO and natural enstatite with 6.43 per cent FeO were ground together to a very fine powder and subjected to 15,000 lb/in² pressure of water vapor at 1000°C . The powder remained a powder. There was no indication of the formation of a liquid.

Improvement of the hydrothermal quenching apparatus. The hydrothermal quenching apparatus (Tuttle) has been improved by using stellite instead of stainless steel as the material of the pressure chamber and also for the plungers by means of which the confining pressure is applied. The greater strength of stellite at high temperatures has extended the temperature range to which experiments can be carried at any given pressure, and in particular has made it practicable to investigate at higher temperatures than heretofore the possibility of the formation of serpentine liquid.

THE SYSTEM SODA—SILICA—WATER

Further work has been done on the system $\text{Na}_2\text{O}—\text{SiO}_2—\text{H}_2\text{O}$, which consti-

tutes a problem of great complexity. According to the present method of study (Morey), glass or crystalline material of the desired composition is placed in a closed vessel, heated to the desired constant temperature, and water pumped in until the desired pressure is reached. The water becomes vaporized in the hot pressure vessel and the pressure is kept constant by a regulator. After removal from the furnace the condensed vapor and the separate silicate-rich liquid are analyzed, and if any crystals are present they are identified by means of the polarizing microscope. Analysis of the vapor is necessary because it contains significant amounts of both soda and silica, and in proportions differing somewhat from the starting material. For example, with sodium disilicate as starting material at 400° C and 1000 atmospheres pressure, all is dissolved in the vapor until 0.9 g have been added, whereupon the first trace of liquid appears. The vapor now contains 5.5 per cent of sodium disilicate. This represents the vapor solubility of sodium disilicate at 400° temperature and 1000 atmospheres pressure. The first trace of liquid formed contains over 70 per cent of silica, instead of the 66 per cent put in as disilicate.

As the amount of disilicate present is increased, the amount of liquid increases until crystalline disilicate appears. Vapor, liquid, and crystals then coexist at this temperature and pressure. The vapor now contains about 1.38 g dissolved material in a volume of 13 cc, and much more soda than corresponds to disilicate composition; and the actual amount of silica is also greatly increased over that corresponding to the vapor solubility of disilicate itself.

From one point of view the phenomenon of the solubility of nonvolatile solid material in the vapor is not a strange one. The water, when above its critical point

and hence a gas, is highly compressed. At 400° C and 1000 atmospheres pressure the density of water is 0.71, not greatly different from that of water under ordinary conditions. That it should have a significant solvent power is to be expected on physical-chemical grounds, and is in harmony with geological observations. Indeed, the transport of nonvolatile mineral components by means of a highly compressed aqueous vapor may very well be an important method of mineral formation and ore deposition; in a different field the phenomenon is frequently observed in high-pressure steam turbines, where the deposition of quartz on the turbine blades diminishes efficiency.

The solubility curves in the system $\text{H}_2\text{O}-\text{Na}_2\text{O}-\text{SiO}_2$, both vapor solubility and liquid solubility, are being determined by change in the proportions of silica, and by change in the pressure, at constant temperature, and for a series of temperatures. The study is greatly complicated by the occurrence of what is termed a "critical" region which includes the disilicate region but which does not extend to the region in which quartz crystallizes. The phenomena met with are of a type never studied heretofore, and they require careful theoretical investigation. Progress is slow, but it is important that both experimental and theoretical investigations be carried out in a comprehensive way because of the fundamental basis they afford for all work which includes nonvolatile constituents together with volatile constituents above their critical temperatures.

HIGH-PRESSURE STEAM FILTER AUTOCLAVE

The autoclave mentioned in last year's report is now in operation. Difficulties with leaks have been obviated, and various improvements, including a complete redesign of the connection between the auto-

clave and the steam boiler, have been made. Two heated valves were arranged in such a way that many of the connections previously made by means of separate tees and crosses are now made within the bodies of the valves, and many joints are eliminated that were potential sources of leaks. The new arrangement provides great accessibility to the remaining joints, so that it is easier to find a leak when one does occur.

When the equipment had been re-assembled in its improved form, a filtrate was obtained after a few preliminary trials. This first filtrate, which was obtained at 300°C under 50 bars pressure of steam in the part of the $\text{K}_2\text{O}-\text{SiO}_2-\text{H}_2\text{O}$ system where KHSi_2O_5 is a solid phase, was found by chemical analysis to contain excess water caused by condensation of the steam during cooling. This was not an unexpected complication, for there are very few a priori considerations on which to base the choice of cooling schedule. This difficulty is to be overcome by exploring the effect of different cooling schedules on the water content of filtrates obtained under the same conditions as the first one.

The preliminary trials already made have established one important characteristic of the apparatus, namely, that equilibrium is established quickly because the reaction mixture is stirred. Four hours were sufficient to cause complete reaction between $\text{K}_2\text{Si}_2\text{O}_5$, quartz, and water to form KHSi_2O_5 and a liquid that became a filtrate. The minimum time for the attainment of equilibrium has not yet been determined. The stirrer now in use is a silver one made in accordance with specifications based on the results of the trials of brass models. Its performance has been satisfactory so far.

In preparation for systematic work with this new apparatus, with which we expect

to make precise measurements on the composition of liquid phases in alkali-silica-water systems, a number of potassium silicates and their known hydrates have been synthesized (Prodan) and X-ray spectrographs have been taken for use as comparison standards in the identification of solid phases that may be obtained in the course of solubility determinations.

EXPERIMENTS ON THE FORMATION OF ANALCITE

Analcite is an important mineral with the composition $\text{Na}_2\text{O} \cdot \text{Al}_2\text{O}_3 \cdot 4\text{SiO}_2 \cdot 2\text{H}_2\text{O}$, which may be thought of as that of jadeite with the addition of two molecules of water. Beginning before the war, a series of experiments was carried out by subjecting an artificial zeolite of the composition $\text{Na}_2\text{O} \cdot \text{Al}_2\text{O}_3 \cdot 5\text{SiO}_2$ to moderate temperatures in the presence of water under pressure in an effort to determine whether or not analcite is formed below a certain temperature and pressure, and albite above that point (Morey). Some of the experiments illustrate the difficulty of being certain as to true equilibrium relations in some silicate mixtures. For example, in a recent test made at a pressure of $12,000\text{ lb/in}^2$ three different materials in separate containers were placed in the pressure vessel at the same time. The first, which was the artificial zeolite, produced albite; the second, a glass of jadeite composition, gave beautiful crystals of analcite together with a small amount of albite; and the third, a sample of natural analcite, at the end of the experiment, which ran for $2\frac{1}{2}$ days, was found to be about half decomposed with the formation of crystalline albite. An additional run for 13 days at $14,200\text{ lb/in}^2$ and 390°C gave the following results: natural analcite was unaltered; glass of jadeite composition produced analcite; the synthetic zeolite gave a mixture of glass

and albite not well crystallized; and natural crystalline jadeite was unaltered.

On the other hand, experiments with the new hydrothermal quenching apparatus on glass of jadeite composition in the presence of water seem to establish a definite temperature-pressure equilibrium curve below which analcite is formed, and above which only an incompletely crystallized mixture of nepheline and albite results (Tuttle, Yoder). A temperature of 540°C and a pressure of $16,000\text{ lb/in}^2$ represent a typical point on the curve. It is interesting to note that with natural jadeite as a starting material the conversion to nepheline and albite is complete.

HIGH PRESSURES COMBINED WITH HIGH TEMPERATURES

The above-mentioned observations relate closely to the problem of the stability of jadeite, which is believed by many petrologists to form only under conditions of elevated pressure. As a part of the basic program of the Laboratory, according to which we seek to understand the important petrologic processes, we plan to investigate not only the behavior of silicate mixtures in the presence of water subjected to pressures sufficiently high to prevent the volatile material from escaping, but also the effects of still higher pressures on the properties and formation of minerals. Accordingly, experiments have been started (Yoder, Adams) by the use of an apparatus which contains a small sample of material under a directed pressure of 5000 atmospheres or higher and heated externally to a temperature of several hundred degrees. This experimentation puts

a high premium on materials that can withstand high stress at temperatures of 700 or 800°C , or more. New types of steel and other alloys developed during the war period have greatly increased the prospects of success with this kind of high-pressure experimentation.

Several methods of attack on the jadeite problem are being used. These include studies on the breakdown of natural jadeite with and without the presence of water; long heating at various temperatures of synthetic glass of the desired composition and of compressed powders of various natural and synthetic materials mixed in proper proportion to give the composition of jadeite; sintering powders under conditions of high hydrostatic pressure and moderate temperatures; heating glass of jadeite composition with various fluxes; hydrothermal treatment of glass at moderate pressures and moderate to high temperatures; heating glass in closed silica tubes with sodium fluoride, water, or any suitable "mineralizer." Other methods of attack such as the investigation of the reactions of colloiddally activated materials are being given consideration.

At the time this report is being prepared, plans are also being made for the construction of an internally heated pressure vessel with which it is hoped to attain pressures of 10,000 atmospheres or more combined with temperatures up to 1200 to 1400° and perhaps as high as 1500°C . The purpose of this part of the work will be not only to enlarge our perspective on the whole subject of high-pressure effects, but also to provide another approach to the determination of certain thermal properties of rock-forming minerals.

ANHYDROUS SILICATE MIXTURES

THE SYSTEM POTASH—MAGNESIA—
ALUMINA—SILICA

As was pointed out in a previous report, a knowledge of the phase equilibrium relations between liquids and crystals in the "dry" quaternary system K_2O — MgO — Al_2O_3 — SiO_2 is necessary before H_2O can be included as an additional component for the purpose of attacking the problem of the stability and crystallization relations of the dark micas (biotite, phlogopite, etc.) in igneous rocks. During the past year very substantial progress has been made on the arduous task of working out this quaternary system.

In a phase equilibrium study of such a system, one of the first essentials is a knowledge of the relations in the four limiting ternary systems. The relations of MgO — Al_2O_3 — SiO_2 were reported from this Laboratory some years ago by Rankin and Merwin, and recently Schairer and Bowen reported the relations for K_2O — Al_2O_3 — SiO_2 . The system K_2O — Al_2O_3 — MgO is in itself of very little interest to us because such compositions have no geological importance, and all the temperatures involved are very high. A knowledge of the limiting ternary system K_2O — MgO — SiO_2 , however, is of prime importance in establishing the relations in the quaternary system. No information on the phase relations in this system was available previously; and, therefore, one of the first steps was to obtain these data.

The system K_2O — MgO — SiO_2 (Roedder). During the past year a study of this very interesting ternary system has been completed, and the data are being assembled for publication. Mr. Edwin W. Roedder, of Columbia University, working under a fellowship grant by the Institution, completed this work at the Geophysical Laboratory under the direction of

Schairer. The results of this study will be offered as a Ph.D. thesis to the University Graduate School.

In order to establish the phase relations between liquids and crystals in this system, 134 separate compositions were prepared and studied by the method of quenching. The compositions and temperatures at the ten binary invariant points (nine of them eutectics) and at the thirteen ternary invariant points (seven of them eutectics) within the ternary system were determined. The data show nine binary systems and only three ternary systems within the system K_2O — MgO — SiO_2 .

Because of the high viscosity of the melts and the sluggishness of crystallization and inversion, no quartz was encountered in the fields of silica at appropriate temperatures. For this reason, it was not possible to locate the ternary invariant point with quartz, tridymite, $K_2O \cdot MgO \cdot 5SiO_2$, and liquid as the coexisting phases; and the metastable ternary point, a eutectic, with tridymite, potassium tetrasilicate, $K_2O \cdot MgO \cdot 5SiO_2$, and liquid was located instead of the stable point for which quartz would take the place of tridymite.

Fields of stability of eight solid crystalline phases previously encountered in the limiting binary systems were delineated in the ternary system, viz., cristobalite, tridymite, forsterite, clinoenstatite, periclase, potassium tetrasilicate, potassium disilicate, and potassium metasilicate. The optical properties and X-ray patterns for these solid phases agree with those previously reported. Four new ternary compounds were found and their temperatures, stability relations, and fields of stability established. $K_2O \cdot 5MgO \cdot 12SiO_2$ forms hexagonal crystals and has an incongruent melting point at $1174 \pm 2^\circ C$. $K_2O \cdot MgO \cdot 5SiO_2$ has a congruent melting point at

$1089 \pm 2^\circ \text{C}$ and forms isometric crystals which invert at a lower temperature to a twinned nonisotropic form. X-ray data show that it is structurally nearly identical with leucite ($\text{K}_2\text{O} \cdot \text{Al}_2\text{O}_3 \cdot 4\text{SiO}_2$), which also has an isometric high-temperature form and a similar inversion. $\text{K}_2\text{O} \cdot \text{MgO} \cdot 3\text{SiO}_2$ forms hexagonal (possibly pseudo-hexagonal) crystals and has a congruent melting point at $1134 \pm 2^\circ \text{C}$. X-ray data indicate that it is similar to kalsilite, one of the crystalline forms of $\text{K}_2\text{O} \cdot \text{Al}_2\text{O}_3 \cdot 2\text{SiO}_2$. $\text{K}_2\text{O} \cdot \text{MgO} \cdot \text{SiO}_2$ forms isometric crystals and has a melting point, probably congruent, well above 1600°C .

In the potash-rich region of the ternary system near the compositions of $\text{K}_2\text{O} \cdot \text{MgO} \cdot \text{SiO}_2$ and $\text{K}_2\text{O} \cdot \text{SiO}_2$ some difficulty was encountered on account of the loss of K_2O by volatilization from the melts, which have high liquidus temperatures. In this region it was necessary to make a chemical analysis of each sample held at the liquidus temperature. No difficulties with potash volatilization were encountered in the more siliceous regions of the ternary system.

Method of attack on the quaternary system. A quaternary silicate system, that is, all possible mixtures of the four components, may be represented most conveniently by a regular tetrahedron, each apex of which represents 100 per cent of one component. The four faces of this tetrahedron are equilateral triangles on which may be plotted the phase equilibrium data for the four limiting ternary systems. Each of these limiting systems is divided into fields (areas where one solid phase is in equilibrium with liquids whose composition lies within the area) of stability of the several solid phases which appear on the liquidus surface. The liquidus surface is the temperature surface where the last crystal dissolves on heating or, conversely, where the first

crystal appears on the cooling of a homogeneous liquid. In each of the limiting ternary systems the fields are delineated by boundary curves along which two solid phases are in equilibrium with liquids whose compositions lie in the boundary curve. These boundary curves in the limiting ternary systems become curved surfaces (quaternary divariant surfaces with two solid phases in equilibrium with liquids whose compositions lie in such a surface) within the tetrahedron.

In the limiting ternary systems, three boundary curves intersect at a ternary invariant point, at a particular temperature, with three solid phases in equilibrium with only that particular liquid whose composition is represented by the ternary point. Each such ternary invariant point becomes a univariant line (a curved line with three solid phases in equilibrium with a liquid whose composition lies in the line) within the tetrahedron. Three curved surfaces (faces of primary phase volume) intersect within the tetrahedron to form such a curved line. Four such univariant lines intersect within the tetrahedron at a quaternary invariant point, at a particular temperature, with four solid phases in equilibrium with only that particular liquid whose composition is represented by the point. At such a quaternary invariant point, six curved surfaces (from four adjacent primary phase volumes) intersect in a point.

Thus it is seen that fields (areas) of stability in the ternary triangle with one solid phase become primary phase volumes in the tetrahedron; that boundary curves (with two solid phases) become curved surfaces defining primary phase volumes within the tetrahedron; and that ternary invariant points (with three solid phases) become univariant lines within the tetrahedron. In other words, the tetrahedron is made up of a series of interlocking

primary phase volumes (polyfaced geometrical figures with curved faces), each such primary phase volume representing the compositions of all quaternary liquids in equilibrium with only one solid phase. Where two primary phase volumes have a face in common, the compositions of all quaternary liquids in equilibrium with these two solid phases lie in this common face (a divariant curved surface). Where three primary phase volumes join, they have a univariant line in common. In such a curved line are the compositions of all quaternary liquids in equilibrium with (saturated with respect to) these four solid phases, that is, a quaternary invariant point.

The study of the equilibrium relations within the quaternary system consists in (1) delineating the portion of the tetrahedron occupied by each primary phase volume, (2) determining the compositions and temperatures along the edges and at all apexes of each primary phase volume, (3) determining the mutual relations within the tetrahedron of the many primary phase volumes involved, and (4) finding the location of a sufficient number of isotherms to define as closely as desired the liquidus and solidus temperatures within the system.

If compositions for study were selected at random within the tetrahedron, it would take an impossibly large number of points to define the system. One method of attack would have been the study of compositions in a series of planes parallel to one face of the tetrahedron. We have chosen, however, to initiate our study by selecting a series of triangular planes, called joins, that have certain minerals, oxides, or compounds as apexes of the joins. By a judicious selection of joins, the maximum of information may be obtained with a minimum number of compositions studied. By our study of these selected planes (joins), we are, in effect,

locating the positions and shapes of primary phase volumes and therefore the approximate positions of quaternary invariant points, because we thus discover which primary phase volumes are cut by a given join and which univariant lines pierce it. We have also determined the temperature relations in the primary phase volumes, along divariant boundary surfaces, and in the univariant lines lying in the plane of the join, and we can set rough limits for the temperatures of the important quaternary invariant points.

The join leucite—forsterite—silica (Schairer, Chappellear). During the past year this join has been completed except for a few long runs (in viscous compositions near the leucite-silica side of the join) which require quenching runs of long duration to attain equilibrium. Forty-five compositions were prepared and studied to define the relations in this join. The plane leucite—forsterite—silica is a ternary system within the quaternary system. Therefore, it is a composition plane which locates a temperature maximum in each quaternary univariant line that pierces this plane; and it thus effectively partitions the quaternary system. During crystallization, whether it be equilibrium crystallization or fractional, no liquid can cross this composition barrier. There are three piercing points in this join. They are the three ternary invariant points, two of them reaction points (forsterite + clinoenstatite + leucite + liquid and clinoenstatite + leucite + potash feldspar + liquid) and one a eutectic (clinoenstatite + potash feldspar + tridymite + liquid). Since these three ternary invariant points are temperature maxima on three quaternary univariant lines which continue within the tetrahedron on both sides of the plane toward six quaternary invariant points, they set an upper temperature limit for these six important quaternary points. The fields of

forsterite, clinoenstatite, cristobalite, tridymite, leucite, and potash feldspar in this join are cross sections of primary phase volumes in this plane through the tetrahedron. The interrelation of the fields defines the interrelation of the several primary phase volumes. Isotherms in the fields are traces of isothermal curved surfaces in the primary phase volumes.

Sufficient progress has been made in the experimental study of silicate systems to indicate the nature of the final residual liquids from the fractional crystallization of complex rock-forming magmas. The ternary system leucite—forsterite—silica illustrates in a striking manner the direction of change of composition in silicate liquids during crystallization. The femic constituents (in this case the olivine, forsterite, and the pyroxene, clinoenstatite) are almost completely removed first, leaving residual liquids that are essentially potash-alumina silicates. Appropriate compositions in the system, even though there is less than 0.5 per cent forsterite in their total composition, crystallize the femic mineral clinoenstatite first and yield a residual liquid almost quantitatively free from the femic constituent.

The join leucite—forsterite—potassium disilicate (Schairer, Chappellear). This join is a triangular plane within the tetrahedron which represents the quaternary system. It has one edge in common with the join leucite—forsterite—silica. This edge is the binary system leucite—forsterite. Another edge, leucite—potassium disilicate, is a binary system within the ternary system $K_2O-Al_2O_3-SiO_2$, one of the faces of the tetrahedron. The third edge is forsterite—potassium disilicate, a join (not binary) in the $K_2O-MgO-SiO_2$ face of the tetrahedron. The join forsterite—potassium disilicate crosses the fields of forsterite, potassium disilicate, and $K_2O \cdot MgO \cdot 3SiO_2$ (whose composition lies neither in this

join nor in the triangular join leucite—forsterite—potassium disilicate).

During the past year the liquidus determinations have been obtained for the join leucite—forsterite—potassium disilicate, the fields in the join have been delineated, and the two piercing points of univariant lines have been located as to composition and temperature. Thirty-eight compositions in this join were prepared and studied. The most noteworthy feature of this join is a continuous rise in temperature from the forsterite—potassium disilicate edge in the field of $K_2O \cdot MgO \cdot 3SiO_2$ to the temperature $1265 \pm 5^\circ C$ at the piercing point where forsterite and leucite coexist as solid phases with the compound. This temperature is far above the congruent melting point of $K_2O \cdot MgO \cdot 3SiO_2$ at $1134 \pm 2^\circ C$ in the ternary system $K_2O-MgO-SiO_2$. As noted above, the compound $K_2O \cdot MgO \cdot 3SiO_2$ and crystals obtained from compositions in its field in the join leucite—forsterite—potassium disilicate are structurally the same and are similar in structure to kalsilite, one of the forms of $K_2O \cdot Al_2O_3 \cdot 2SiO_2$. This field is probably the field of kalsilite— $K_2O \cdot MgO \cdot 3SiO_2$ solid solutions, and more work will be necessary before we can define these solid solutions in the quaternary system. The two piercing points in the join are leucite + potassium disilicate + solid solution + liquid and leucite + forsterite + solid solution + liquid.

The join leucite—clinoenstatite—potassium tetrasilicate (Schairer, Chappellear). This join was chosen to bisect roughly the tetrahedron (not a regular tetrahedron and not quaternary because not all its faces are ternary) formed by the joins leucite—forsterite—silica, leucite—forsterite—potassium disilicate, and parts of two of the faces of the regular tetrahedron which represents the quaternary system $K_2O-MgO-Al_2O_3-SiO_2$. It was chosen so as

to include in one of its edges the compound $K_2O \cdot MgO \cdot 5SiO_2$ which presented some difficulties during the studies of the ternary system $K_2O-MgO-SiO_2$, because this compound apparently had two crystalline forms whose mutual relations were not easy to understand.

Forty-three compositions have been prepared, and quenching data on these are completed except for five which have proved difficult to crystallize. Furthermore, the preparation of a few additional compositions will be necessary. The outstanding feature of this join is the relation between leucite ($K_2O \cdot Al_2O_3 \cdot 4SiO_2$) and the compound $K_2O \cdot MgO \cdot SiO_2$. The system leucite- $K_2O \cdot MgO \cdot 5SiO_2$ is binary with a complete series of solid solutions and a minimum on the liquidus surface. Leucite and $K_2O \cdot MgO \cdot 5SiO_2$ are structurally nearly identical, and the leucite- $K_2O \cdot MgO \cdot 5SiO_2$ solid solutions show the same structure. Each of the compounds has a high-temperature isometric form which inverts at some lower temperature ($603^\circ C$ in the case of leucite) to a birefringent nonisotropic form. The inversion does not always occur during quenching, and is dependent on the composition, size of crystals, and rate of cooling. With the obtaining of this bit of knowledge all the apparent discrepancies in the early observations disappeared.

The join leucite-clinoenstatite-potassium tetrasilicate shows a large field of forsterite, a very small field of the hexagonal compound $K_2O \cdot 5MgO \cdot 12SiO_2$, a large field of leucite- $K_2O \cdot MgO \cdot 5SiO_2$ solid solutions, and small fields of potassium tetrasilicate and potash feldspar. There are two piercing points of univariant lines in the join. One of them, forsterite + solid solution + $K_2O \cdot 5MgO \cdot 12SiO_2$ + liquid, has been located as to temperature and composition. The other, potassium tetrasilicate + potash feldspar + solid

solution + liquid, has so far been only roughly located owing to difficulties with crystallization in this low-temperature region of the join.

The join cordierite-forsterite-leucite (Schairer, Chappellear). This join was chosen as the first exploratory plane in that part of the tetrahedron which lies between the join leucite-forsterite-silica and the face $MgO-Al_2O_3-SiO_2$. One edge of this join, the binary system leucite-forsterite, is common also to the joins leucite-forsterite-silica and leucite-forsterite-potassium disilicate already discussed. Forty compositions were prepared during this year and studies of all of them have been completed. Only a few more compositions will be necessary to locate more accurately the boundary surfaces or isotherms in the join. The fields of forsterite and spinel occupy a large portion of this join. There is a small field of leucite and there are very small fields of cordierite and mullite. None of the liquidus temperatures is below $1425^\circ C$, and equilibrium is attained rapidly. There are only two piercing points of univariant lines in this join, leucite + forsterite + spinel + liquid and cordierite + spinel + mullite + liquid. Both have been located accurately as to temperature and composition.

The join cordierite-leucite-silica (Schairer, Chappellear). This join continues the exploration in that part of the tetrahedron which lies between the join leucite-forsterite-silica and the face $MgO-Al_2O_3-SiO_2$. It completes a smaller tetrahedron (not a regular tetrahedron, and not quaternary because not all its faces are ternary) within the regular tetrahedron which represents the quaternary system $K_2O-MgO-Al_2O_3-SiO_2$, and it was chosen because it should define the relations between the important minerals cordierite, tridymite, potash feldspar, leucite,

and spinel. Forty-nine compositions have been prepared and are now being studied. Quenching experiments already completed approximately outline the relations in the join, but a few additional samples must be prepared in order to complete this join, and they will probably all involve difficulties in crystallization and length of run because of the viscosity of melts in the region where the fields potash feldspar, leucite, and cordierite come together.

The field of cordierite occupies a large region in the center of this join; the fields of mullite, spinel, leucite, cristobalite, and tridymite are considerably smaller; and the field of potash feldspar is a very small one lying close to the leucite-silica edge near its middle. There are five piercing points of univariant lines in this join. Two of them have been located accurately as to temperature and composition: mullite + spinel + cordierite + liquid, and spinel + cordierite + leucite + liquid. The three others have been located only approximately by the experiments that are still in progress: mullite + cordierite + tridymite + liquid, cordierite + tridymite + potash feldspar + liquid, and leucite + potash feldspar + cordierite + liquid.

Summary and plans for further work on this system. Gratifying progress has been made on the quaternary system $K_2O-MgO-Al_2O_3-SiO_2$ in the short span of one year. A large amount of the information has been extracted from studies of only 215 quaternary compositions; and during the next year we hope to complete the joins leucite-clinoenstatite-potassium tetrasilicate and cordierite-leucite-silica. All necessary data on leucite-forsterite-silica, leucite-forsterite-potassium disilicate, and cordierite-forsterite-leucite should be in hand after a few weeks' further work. The data on these joins together with the data on one or both of

the other two mentioned above will be assembled for publication. Work on several additional carefully selected joins will be started during the course of next year.

THE QUATERNARY SYSTEM MAGNESIA— FERROUS OXIDE—ALUMINA—SILICA

During the past year a start has been made on the system $MgO-FeO-Al_2O_3-SiO_2$ (Keith). No data were available on the mutual melting and stability relations in this very interesting system, which includes the cordierites; the magnesium-iron spinels, olivines, and pyroxenes (including aluminous pyroxenes, if any); the magnesium-iron garnets; the magnesio-wüstites; mullite; corundum; periclase; tridymite; and cristobalite, nearly all of which occur as rock-forming minerals in the igneous or metamorphic rocks.

Since little or no information was available on the ranges of composition, on the stability relations, or on the optical properties of the magnesium-iron cordierites, the join cordierite ($2MgO \cdot 2Al_2O_3 \cdot 5SiO_2$)—iron-cordierite ($2FeO \cdot 2Al_2O_3 \cdot 5SiO_2$)—FeO was selected for study. The compositions of both these cordierites lie in the primary phase volume of mullite (i.e., they melt incongruently); and, consequently, these cordierites appear as solid crystalline phases only at sub-liquidus temperatures. The liquidus relations in the join were completed, and sub-liquidus studies are still in progress. A study of several additional joins will be necessary before an adequate picture of the relations of the two cordierites to each other and to the pyroxenes, olivines, spinels, and garnets can be obtained.

THE SYSTEM SODA—BORIC OXIDE—SILICA

Work on the difficult system $Na_2O-B_2O_3-SiO_2$, which was interrupted by the war, has been resumed in an effort to extend our knowledge of the behavior of

mixtures containing B_2O_3 , which in combination with SiO_2 and other materials shows a number of striking peculiarities. One serious obstacle to completing the research has been the difficulty of obtaining crystallization in some of the mixtures in the central part of the ternary diagram. In this region all the usual tricks that have been employed in past years to deal with difficultly crystallizable mixtures have been of no avail. Of the 92 new mixtures that have been studied, however, the majority were obtained in crystalline form. Recently success with some of the others has been achieved by heating the mixtures in the presence of water under pressure. In the difficultly crystallizable region a new compound has appeared and has been tentatively assigned the composition $Na_2O \cdot$

$B_2O_3 \cdot 2SiO_2$, analogous to the mineral danburite. The new compound has a sharp congruent melting point at about $740^\circ C$.

This compound is of considerable interest. It resembles danburite in having a much higher refractive index than would be expected from its composition. Glass in either system of the composition of the compound has a refractive index near 1.51. That of danburite is ± 1.63 , and this new compound is isotropic, n being a little greater than 1.58. In each case there is an unusually great condensation on crystallizing.

Experimentation on this system is being continued with the expectation that a well rounded contribution can be made at an early date.

STUDIES ON THE INVERSION OF QUARTZ

Samples of powdered quartz with a thermocouple inserted in the sample were heated in a platinum crucible in such a manner that the inversion temperature was approached at the uniform rate of $1^\circ C$ per minute (Tuttle). The absorption of heat at the inversion caused a break in the heating curve which accurately locates the inversion temperature.

The inversion temperature for 14 quartz specimens from localities representing greatly different geologic environments has been determined with respect to an arbitrarily chosen reference quartz. The limit of error is believed to be within $\pm 0.05^\circ C$.

Quartz from lavas (suggesting high temperature of formation) inverted to the high-temperature modification at lower temperatures than quartz grown from aqueous solutions (suggesting low temperature of formation). The greatest difference observed in the specimens examined was $1.89^\circ C$. Of the specimens of quartz avail-

able for study, the one with the presumably lowest formation temperature was a novaculite from Arkansas, which gave the highest inversion temperature yet measured— $0.69^\circ C$ higher than the arbitrary reference quartz. On the other hand, 4 specimens of quartz from lavas inverted 0.58 – $0.69^\circ C$ lower than the reference quartz. Between these two types of quartz are, in order of decreasing inversion temperature: quartz from Herkimer County, New York, which had grown in vugs in limestone; vein quartz from Washington, D. C.; quartz from Hot Springs, Arkansas; quartz from Minas Geraes, Brazil (reference); quartz from a tuff from New Mexico; and quartz from a graphic granite from Maine.

This sequence is roughly the order of crystallization temperature as deduced from geological environment. It appears, then, that the inversion temperature is inversely related to the temperature of formation.

Theoretical considerations suggest a reason for this variation of the inversion temperature with temperature of formation. High quartz has a greater specific volume than low quartz, and, consequently, a somewhat more open structure. This open structure permits a greater amount of so-called impurities to enter into solid solution in crystals which have formed above the inversion temperature. Thermodynamic theory shows that in general an inversion temperature is lowered when the impurities have the greater solubility in the high-temperature modification. It is suspected that the solid solution is largely of the interstitial type, although

the occurrence of substitution appears to be possible.

The need for better "geologic thermometers" is very real. For example, the controversy on the origin of granite could probably be minimized if the temperature of formation of the different types of granite could be ascertained. Light could also be thrown on such problems as the origin of graphic granite, that of quartz veins, and that of pegmatites.

It is planned to continue measuring the inversion temperature of natural quartz from various localities, especially those localities where the relative temperature of formation can be deduced.

STUDIES ON VOLCANIC PRODUCTS

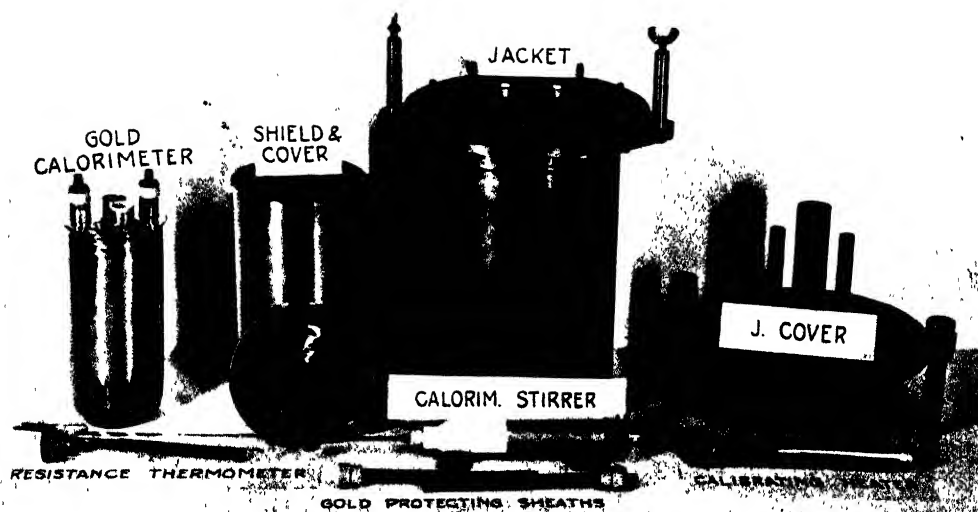
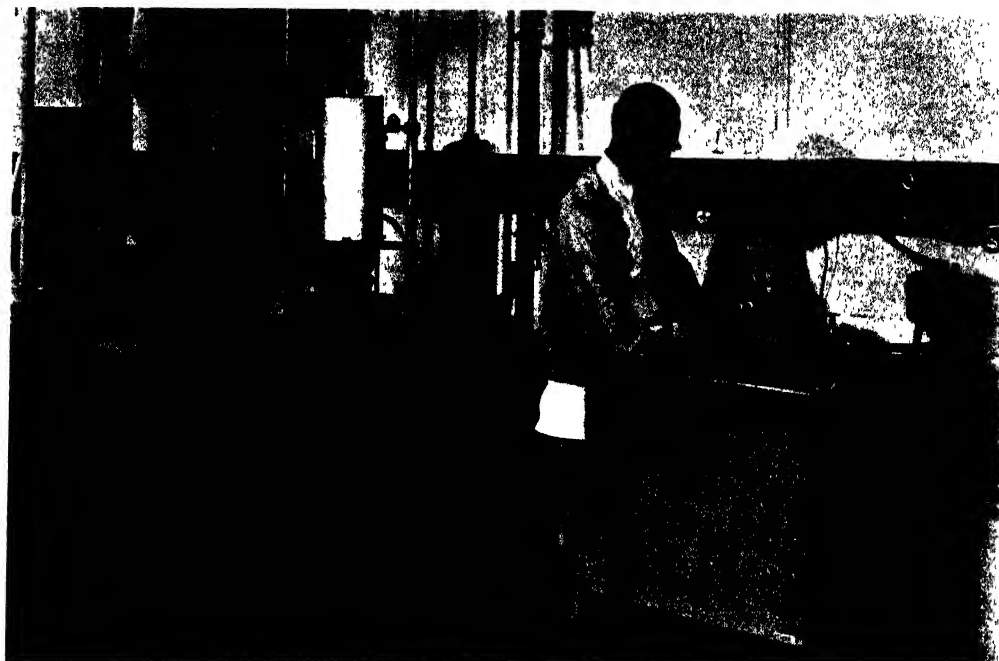
It was stated in last year's report that it seemed preferable for the immediate future to confine the work on volcanic products to the study of samples collected on the various expeditions to Santiaguito, the new volcanic edifice on the old volcano of Santa Maria in Guatemala. This work has been continued (Zies, Merwin, Kaltenthaler), and special emphasis has been placed on a thorough analysis of the rocks extruded during and prior to the year 1940. It was deemed advisable to concentrate our efforts along this line, first, because we wish to know what type of volcanic rock was brought to the surface, and, second, because the interesting array of volatile acid constituents must have been derived from the hot magma while it was on its way to the surface. It has been found that the cold lava still retains some of the volatile materials. This can readily be shown by analysis of the products given off when the lava is heated to about 850° C.

In 1940, hot lava (observed temperature about 725° C) was emerging from the western rim of Santiaguito. This region is very difficult to approach on account of

the rugged terrain and the avalanching of rocks. It was, however, found possible to enter this area and to collect samples of rock only a few minutes after extrusion. This was quite fortunate, because the comparison of the results of the analysis of these rocks with those obtained from the older ones gave a clue to what was probably happening below the surface.

All the rock samples may be designated as andesites. The petrographic analysis of the samples indicated (1) that they contained hypersthene, amphibole, feldspar, and relatively large amounts of glass (25 per cent or more); (2) that they were of widely varying degrees of porosity; and (3) that the more porous ones had the appearance of having been "puffed up" by escape of volatile materials.

The results of the chemical analyses verify and supplement the petrographic analysis. The rocks fall into two groups of andesite, one of which contains about 64 per cent SiO_2 and the other about 57 per cent. It is of interest to note that the ratio of ferrous to ferric iron is by no means constant within either group, even though



The lower picture shows the component parts of the calorimeter used for measuring the heats of solution of silicate minerals in hydrofluoric acid for the purpose of determining latent heats and other thermochemical quantities.

The upper picture shows on the right the assembled heat-of-solution calorimeter in its thermostat, and on the left a partly completed assembly for measuring the specific heats of minerals.

the silica content within each group varies but little.

The petrographic study also revealed that the rocks of either group in which the ratio of ferrous to ferric iron was relatively high contained a mineral that had the optical properties of kaersutite, a titanium-rich (about 10 per cent TiO_2) amphibole. This mineral was especially abundant in the group having the lower silica content. About 25 mg of the pure mineral was isolated and analyzed. It contained only 1.6 per cent TiO_2 . It is obvious that true kaersutite is not present in the Santiaguito lava. A greenish-brown amphibole is also prevalent in our rocks. Other workers have found that when some green varieties are heated to about 850°C they acquire optical properties similar to those of kaersutite. This phenomenon is attributed to the oxidation of the ferrous iron to ferric iron by water in the mineral and the surrounding lava. If this explanation is correct, then it would seem that this process should take place only after the pressure on the magma has been relieved to a point at which the state of oxidation of the hornblende is disturbed.

In our chemical studies, emphasis has been placed also on the determination of the minor constituents such as H_2O , Cl, SO_3 , P_2O_5 , and Ba. This work is time-consuming, but it seems essential for the success of future studies on volcanism. The three volatile constituents will probably acquire added interest when the work on the acid gases emitted by the fumaroles

has been completed. The nonvolatile minor constituents will be of interest in connection with studies of rock alteration by fumarolic activity.

All the constituents just referred to are present in small amounts. The H_2O content is low, only about 0.2 per cent; as a matter of fact, it is considerably lower than is usually found in andesitic rocks.

Field, petrographic, and chemical evidence seems to show that at Santiaguito two types of andesite emerge as an exceedingly viscous, partly crystallized mass which has been forced upward by the volatile constituents escaping from solution in the magma, thus giving the lava its "puffed up" appearance. The slowness of its emergence seems to permit varying degrees of oxidation of the iron-bearing silicates.

The significant results of our investigations of the Santiaguito rocks were presented as a preliminary report at the April meeting of the American Geophysical Union. A formal paper containing a full account of our field and laboratory studies together with deductions that seem to account for the origin of the two types of lava will be prepared at an early date.

The work on the acid constituents in the emanations given off by the fumaroles on Santiaguito is still in progress. Exploratory work on incrustations and altered rocks was undertaken in the course of the year. This has materially facilitated the planning of our work on these materials.

THERMAL PROPERTIES OF MINERALS

The gold calorimeter mentioned in last year's report has now been completed and the auxiliary equipment has been set up so as to proceed expeditiously with the measurement of heats of melting and heats of formation by dissolving the materials

in hydrofluoric or hydrochloric acid. (See plate 1.) In view of the inertness to physical change that the silicate minerals commonly show, we shall be mainly dependent upon the solution method for obtaining the required information on latent heats.

The calorimeter for measuring specific heats will be completed soon. This calorimeter is of the copper-block type, that is, it uses a massive block of copper as the thermal mixing device. The block is surrounded by a jacket, the assembly being held in a thermostat maintained at a constant temperature. When a sample of the material to be investigated is dropped into the receiving well of the copper block, the resultant rise in temperature, measured by a sensitive resistance thermometer, gives a measure of the mean heat capacity of the specimen in the temperature range extending from room temperature to the elevated temperature at which the specimen was held.

During the report year Dr. Sahama, working under appointment by the Institution as Visiting Investigator, rounded out the investigations undertaken at the Pacific Experiment Station of the U. S. Bureau of Mines at Berkeley, California, in co-operation with Dr. Torgeson and Dr. Kelley of that station. A paper by Torgeson and Sahama, describing the design of the Bureau of Mines calorimeter and the method of operation, and presenting the results of measurement on magnesium orthosilicate, magnesium metasilicate, and calcium metasilicate, will appear in an early issue of the *Journal of the American Chemical Society*. A series of measurements was made also at this station on the heat of solution of geikielite (magnesium titanate) and ilmenite (ferrous titanate) as a step in evaluating the heat and finally the free energy of the reaction $\text{Fe}_2\text{SiO}_4 + 2\text{MgTiO}_3 = \text{Mg}_2\text{SiO}_4 + 2\text{FeTiO}_3$.

Working at the Geophysical Laboratory, Sahama made a further synthesis of the thermal data obtained for magnesium and

iron titanates, orthosilicates, and metasilicates. The results of the calculations provide information on the stability of ilmenite and geikielite in the presence of olivine or orthopyroxene. Specifically, it is found that the chemical reactions run in the direction of geikielite at high temperatures and ilmenite at low temperatures. The thermodynamic stability of geikielite in the presence of olivine or orthopyroxene (e.g., in dunites) at high temperatures makes probable a more frequent occurrence of geikielite than has heretofore been thought likely.

The thermal measurements on the magnesium and iron orthosilicates and metasilicates have been used to calculate definite values for the energies of the reactions involving silica. The results show a remarkable difference in the stabilities of the two end members of the magnesium iron metasilicate series, and they lead to a satisfactory agreement between the predictions of thermodynamics and the petrological observations relative to the stability relations of enstatite and orthoferrosilite.

In preparation for a comprehensive series of measurements of thermal properties of all the minerals of prime petrological importance, a number of mineral specimens have been obtained from various sources and have been carefully "purified" by magnetic separation and liquid flotation (Sahama, Kracek, Neuvonen). The preparation of samples of other minerals is going forward. Excellent samples of feldspars, feldspathoids, the different modifications of silica, and the different modifications of aluminum metasilicates have been obtained for use as starting materials in the preparation of suitable samples for the contemplated measurements.

OTHER INVESTIGATIONS

STATISTICAL PETROGRAPHY

In general terms, all the problems of descriptive petrography fall into one of two categories: the petrographer is attempting to describe either a single rock type or the relation of two rock types to each other. A successful statistical revamping of petrography will, therefore, require usable numerical definitions, both of rocks in themselves, and of their relations to one another. It will also require methods of measurement suitably precise and economic so that adequate data may be obtained by reasonable expenditure of time. Finally, and perhaps most difficult, techniques designed to yield random samples of rocks must be developed.

Measurements. During the past year some progress has been made along each of the avenues noted above (Chayes). A point counter has been developed which permits comparatively rapid and precise analysis of thin sections. With this instrument, an area of $\frac{3}{4}$ square inch can be analyzed in about 20 minutes, and any number of constituents can be recorded; the reproducibility of the results has been established by considerable experimentation. Well over three hundred analyses have been made with the instrument, a description of which is scheduled for early publication.

Sampling. The approach to the sampling problem has so far been largely intuitive. This problem is complicated by the fact that the best type of petrographic sampling has always been, and probably always will be, consciously "representative," whereas the machinery of statistics functions properly only on samples which are "randomly" taken. In certain cases the definitions may coincide: a statistician may regard a Jones sampler as yielding a random sample, while a petrographer may

hope it will yield a representative one; but the sample will not be affected by this difference of opinion. On the other hand, no petrographer would be likely to bring home an aplite or a black knot as a sample of a quarry granite; whereas a truly random sampling procedure in which each of a large number of quarries was represented by only one specimen would inevitably turn up a few of a peculiar kind.

This matter of sampling will undoubtedly be one of the trouble spots of the whole program. So far, it has been sidestepped by accepting the petrographer's definition in taking the sample and the statistician's in deciding what to study in it. A group of attributes is set up for use in sampling; the variables chosen for study are such as will be moderately affected, if at all, by the fact that specimens not showing these attributes are eliminated from the sample. Thus, the fact that out of 150 samples of granite from all over New England not one contains as much as 40 per cent quartz, and very few carry over 37 per cent, provides a reasonable basis for inference about the maximum quartz content of granite even though the sample is restricted to specimens containing not less than 15 per cent quartz. Incidentally, the distribution of quartz in these granites is sensibly normal even though a lower bound to the quartz content is part of the definition of granite.

Definition of a single rock type. The main object of the work on granite is to arrive at an effective numerical definition or description of a single rock type. This is being done on an a posteriori basis. Samples have been collected from most of the quarries in each of the major granite-quarrying areas in New England. The basis for selection is rather involved, but practically it amounts to restricting the

samples to material which would be classed as granite on both commercial and scientific grounds. A thin section of each specimen is etched, stained, and analyzed on the point counter. The work is now about half complete; ultimately it will provide information about the frequency distributions of quartz, plagioclase, potash feldspar, muscovite, and biotite. The results will be subjected to the usual variance analysis; and if this offers no reason for questioning the homogeneity of the sample, covariance will be used to determine the extent of correlation between the various mineral pairs.

Relation between two rock types. For a study of the relation between two rock types, a transition zone between granite and gabbro-diorite in Norfolk County, Massachusetts, has been carefully sampled. It has previously been argued that this transition is "gradational"; and the area about Boston was one of the first in which a large-scale gradational transition between these two rock types was suggested. Although the terms are widely used by all geologists, neither "gradation" nor "gradual" has ever been satisfactorily defined. Under any circumstances, one would expect to find that in the transition zone rocks of intermediate composition were dominant. As a limiting condition, it may be suggested that intermediate rocks ought to be at least as frequent as the end members in this zone; a "rectangular" or "equal likelihood" distribution is, therefore, set up as a minimum a priori description of gradation in this case, and it is shown that in the transition zone rocks of intermediate composition are so rare as to invalidate this description. The sample is incompatible with the hypothesis that the transition between granite and gabbro is here truly gradational. A full report of these findings is in preparation.

Utilization of old data. Finally, a really

satisfactory reorientation of petrography must make as much use as possible of old data. This is probably the hardest part of the job, for the precision of the measurements is rarely known, and information about sampling procedures is scanty or lacking. For these reasons, sample statistics of old data are best regarded as sample descriptions rather than probability statements. The question to be answered here is not whether some particular hypothesis should be accepted or rejected, but merely whether relations required by the hypothesis are actually shown by the sample.

A study of this type has been made of variations in the bulk chemical composition of "rapakivi" granites. According to the dominant hypothesis of the origin of rapakivis, feldspar develops largely or wholly at the expense of quartz. As these two are by far the major constituents, it follows that there ought to be high negative correlation between them. It is easily shown that this is not the case in the Finnish rapakivis, in which quartz and feldspar are virtually uncorrelated. The hypothesis is therefore not an adequate explanation of known variations in bulk chemical composition of the rapakivi granites.

RADIOACTIVITY

A series of measurements of the radium content of meteorites has been made (Davis). It was found that iron meteorites as well as the metallic and silicate portion of pallasites have less than half the amount of radium found in the average ultramafic rock, although some rocks of that type have a radium content as low as that of the lowest meteorites. Tentatively, the average amount of radium in meteorites is considered to be 5×10^{-15} g per gram. This value is probably high rather than low.

In accordance with a suggestion by the Navy Department, samples from six cores obtained from the ocean bottom in the South Atlantic by the U. S. Navy Antarctic Expedition of 1946-1947 were received by the Laboratory and measurements made of their radioactive content at various positions along the cores (Urry). Seventy determinations of radium content were made, and it was found that the curves showing radio content as a function of depth below the ocean bottom are qualitatively smaller than the curves previously obtained from cores from the North Atlantic Ocean.

SEISMIC PROSPECTING OF THE EARTH'S OUTER LAYERS

The Department of Terrestrial Magnetism and the Geophysical Laboratory have been co-operating in a program of seismic prospecting for the exploration of the depths of the earth by means of elastic waves initiated by large charges of high explosive and recorded at various distances from the explosion. Convenient and sensitive portable instruments have been developed for this kind of work, and already a sufficient number of records have been

obtained to permit the drawing of interesting conclusions concerning the crustal layers of the earth. Full details concerning the progress of this investigation will be found in the report of the Department of Terrestrial Magnetism.

The following is a list of the papers published during the report year in technical journals. In addition there are several papers, as follows, that have been prepared and are awaiting publication. These are: N. L. Bowen and O. F. Tuttle, "The system $\text{MgO}-\text{SiO}_2-\text{H}_2\text{O}$ "; F. Chayes, "A simple point counter for thin section analysis"; F. Chayes, "On ratio correlation in petrography"; O. F. Tuttle, "A new hydrothermal quenching apparatus"; O. F. Tuttle, "Structural petrology of planes of liquid inclusions"; Wm. D. Urry, "The radium content of varved clay and a possible age of the Hartford, Connecticut, deposits"; Wm. D. Urry, "Radioactivity of ocean sediments. VI. Concentrations of the radioelements in marine sediments of the southern hemisphere"; Wm. D. Urry, "Radioactivity of ocean sediments. VII. Rate of deposition of deep-sea sediments"; and A. H. Stone, "On supersonic flow past a slightly yawing cone. II."

SUMMARY OF PUBLISHED WORK

- (1094) Some unsolved problems of geophysics. L. H. Adams. *Trans. Amer. Geophys. Union*, vol. 28, pp. 673-679 (1947).

Address of the retiring president of the American Geophysical Union given at the Twenty-eighth Annual Meeting of the Union, in April 1947.

Attention is called in this paper to six of the particularly baffling problems in geophysics. These are: the origin of mountain chains; the origin of geosynclines; the cause of igneous activity, including volcanism, a group of phenomena that continue to give us striking evidence of a hot and active interior of the earth; the origin of deep-

focus earthquakes, those caused by some still mysterious processes at depths of as much as four hundred miles below the surface; the origin of the earth's magnetic field, about which much is known, but of which the underlying cause is still uncertain; and the temperatures prevailing in the interior of the earth.

- (1095) Radium content of ultramafic igneous rocks. I. Laboratory investigation. Gordon L. Davis. *Amer. Jour. Sci.*, vol. 245, pp. 677-693 (1947).

A co-operative investigation between this Laboratory and the Department of Geology of Princeton University for the purpose of

obtaining data on the radium content of certain ultramafic rocks has resulted in determinations of the radium content of more than thirty rocks, of the constituent minerals of several, and of one meteorite, a pallasite.

Refinement of the vacuum fusion technique used in previous determinations (ocean-bottom core samples) has enabled reasonably accurate determinations to be made for this class of rocks, already known to be low in radium content. The apparatus and refinements are fully described.

The radium content has been related to the degree of serpentinization as measured by the water content of the rocks, those rocks least serpentinized containing the least radium, 0.005×10^{-12} g per gram of rock. An attempt was made to evaluate the probable distribution of radium among the minerals comprising a dunite.

In general, the radium content of the ultramafic rocks measured was found to be considerably lower than previously reported.

(1096) Continuous heating of a hollow cylinder.
George Comenetz. *Quart. Applied Math.*,
vol. 5, pp. 503-510 (1948).

During the wartime investigation of gun erosion it was important to study the heating of gun barrels. For the case of a gun firing steadily, the heat input through the bore surface decreases as the barrel grows hot. This decrease is nearly enough linear with the time so that it was possible to make the simplifying assumption of an infinitely long hollow cylinder supplied continuously with heat through the inner surface at a linear rate. Formulas for the temperatures within the wall of such a cylinder were derived from some formulas previously published by Carslaw and Jaeger which required the development of summation methods for certain infinite series of orthogonal Bessel functions. Besides the application to gun barrels, these formulas may have application to the heating of a tubular furnace, a chimney, or the insulation on a wire carrying an electric current.

Brief consideration was also given to one case of noncontinuous heating of a hollow cylinder, namely, that of a machine-gun

barrel receiving its heat in a series of discrete pulses occurring at equal intervals of time. In this discussion is introduced the concept of "equivalent steady input temperature," which is the time average of the temperature at a point far enough below the surface for the fluctuations that characterize the temperature near the surface to have been damped out. It is shown that a simple mathematical relation exists between this temperature and the actual fluctuating temperature at the surface.

(1097) The system anorthite—leucite—silica.
J. F. Schairer and Norman L. Bowen.
Bull. Soc. géol. de Finlande, vol. 20, pp.
67-87 (1947).

Quenching experiments were made on 69 different ternary compositions in order to determine the liquidus temperatures, the slopes of the liquidus surface, and the fields of stability of the primary crystalline phases with their boundary curves. These data are assembled in tables and presented graphically as the equilibrium diagram. Fields of anorthite, leucite, potash feldspar, tridymite, and cristobalite are shown on the diagram. There are only two ternary invariant points, one of them (leucite + anorthite + potash feldspar + liquid) a reaction point ($1040 \pm 20^\circ \text{C}$) and the other (anorthite + potash feldspar + tridymite + liquid) a eutectic ($950 \pm 20^\circ \text{C}$).

A marked feature of the ternary system is the extension of the field of anorthite from pure anorthite itself nearly down to the binary bounding system leucite—silica. Even with somewhat less than 5 per cent of anorthite in the total composition of a melt, anorthite may be the first crystalline phase to appear on cooling. From the manner in which the field of potash feldspar is confined to compositions in the immediate vicinity of the leucite—silica side of the diagram, it is clear that all compositions in the whole system will have purged themselves almost completely of anorthite before any crystallization of potash feldspar can occur. A complete discussion of crystallization of this system is given, and the refractive indices of the 69

glasses at 25° C are given in the tables and graphically as a diagram with isofracts.

New data are given for the binary system anorthite—silica which place the eutectic between anorthite and silica at $1368 \pm 2^\circ \text{C}$ and at the composition (weight per cent) anorthite 50.5, silica 49.5 per cent. A new equilibrium diagram is given here for the binary system leucite—silica. The data on which it is based will be published at a later time.

- (1098) The systems $\text{K}_2\text{O—ZnO—SiO}_2$, $\text{ZnO—B}_2\text{O}_3\text{—SiO}_2$, and $\text{Zn}_2\text{SiO}_4\text{—Zn}_2\text{GeO}_4$. Earl Ingerson, G. W. Morey, and O. F. Tuttle. *Amer. Jour. Sci.*, vol. 246, pp. 31–40 (1948).

The properties of the forms of zinc orthosilicate have been determined. Incomplete studies of the system $\text{K}_2\text{O—ZnO—SiO}_2$ have indicated the formation of a compound, $\text{K}_2\text{O} \cdot \text{ZnO} \cdot \text{SiO}_2$, and approximate boundaries for the fields of silica, willemite, ZnO , and $\text{K}_2\text{O} \cdot \text{ZnO} \cdot \text{SiO}_2$. In the system $\text{ZnO—B}_2\text{O}_3\text{—SiO}_2$ the compounds $\text{ZnO} \cdot \text{B}_2\text{O}_3$ and $5\text{ZnO} \cdot 2\text{B}_2\text{O}_3$ have been described and their fields approximately outlined, as well as the region of two liquid layers which extends over the larger part of the ternary diagram. The compound Zn_2GeO_4 , which forms a complete series of solid solutions with Zn_2SiO_4 , is described and optical properties are given for it and the solid solutions.

- (1099) Gamma-radiation from granite. William D. Urry. *Phys. Rev.*, vol. 73, no. 6, pp. 596–601 (1948).

The discrepancy between the observed and computed ionization produced by the gamma rays from Quincy granite as measured by Dr. Victor F. Hess, of Fordham University, led to the conclusion that either the measured uranium, thorium, and potassium contents were much too low, or the granite, and perhaps other rocks, emits a penetrating radiation of unknown origin. This investigation, supported by very recent work on the gamma radiation from potassium, has fully accounted for the gamma-ray ionization on the basis of the measured uranium, thorium, and potassium content.

Computation of the expected ionization

was avoided by comparing the gamma-ray ionization from the granite with that produced by adding to the granite amounts of uranium, thorium, and potassium equal to the reported contents. If the reported contents are correct, and there exists no unknown radiation from the granite, the ionization should be exactly doubled. This was found to be the case. The original discrepancy probably arose in the computation of the ionization, since there was found no serious disagreement between the measurements of Hess and those made at this Laboratory.

The idea that extraneous ionization might be produced by a cosmic-ray transition in the mass of rock surrounding the ionization chamber was considered. It was shown that any effect would be negligible by surrounding the chamber with the ultrabasic rock dunite, which has about 0.5 per cent of the content of three elements causing the ionization from granite. The dunite was used for measurement of the background ionization.

- (1100) The granite problem and the method of multiple prejudices. Norman L. Bowen. *In* *Origin of granite*, Geol. Soc. Amer. Mem. 28, pp. 79–90 (1948).

As a result of field observations, geologists ordinarily succeed only in setting up a number of rival hypotheses. To choose between them, not only are more field observations necessary, but every resource of the fundamental sciences should be employed. Some petrologists have attempted to do this, but in spite of their best efforts there is still a wide divergence of views. Such a problem is the origin of granites.

Two divergent views on the origin of granites are current: one that granite is the product of consolidation of a magma (an igneous rock), the other that it is formed by the replacement of pre-existing rocks through the agency of some diffuse medium which induces "granitization" (the product of a metamorphic process). The cleavage is not as sharp as this might indicate. There is probably no exponent of the first view who does not believe that igneous granitic material has occasionally been introduced into and replaced other rocks, but the contention that

most granitic rock has been formed by this process of granitization is rejected. Similarly, there is no proponent of the second view who does not accept the possibility of a little granitic magma on occasion. The problem resolves itself into a question of how much granite is magmatic and how much metamorphic.

In this paper the field and laboratory evidence for "dry" granitization and "wet" granitization is examined and the processes of crystallization in silicate magmas are discussed. A plea is made for less bias and more co-operation between the "drys" and the "wets" to gain a better understanding of granites and their origin.

- (1101) On supersonic flow past a slightly yawing cone. I. A. H. Stone. *Jour. Math. and Phys.*, vol. 27, no. 1, pp. 67-81 (1948).

When a conical-headed projectile moves through the air with supersonic speed it is preceded by a conical shock wave, attached to the solid cone at their common vertex. At the same time an expansion wave originates at the shoulder of the projectile, where the conical surface meets the cylindrical body. At sufficiently high velocities the flow in the region between the shock wave and the expansion wave is locally supersonic and then is independent of conditions outside this region. Hence for convenience one may assume that the conical part of the projectile extends to infinity.

If the direction of motion of the cone coincides with its axis of symmetry, the characteristics of the air flow past it have been known for some years. The present paper, which relates to wartime research, is concerned with the perturbation on this flow produced by a small "yaw" of the cone, that is, the case in which the axis of the cone is slightly inclined with respect to the direction of its motion. For simplicity it is assumed that the cone is moving with constant velocity in a single direction, and that the effects of viscosity, heat transfer, the boundary layer, and skin friction are negligible. It is further assumed that the angle of yaw is constant, that it is small enough so that its square can

be neglected, and that the flow depends continuously on it.

Differential equations were set up and solved for the velocity components, pressure, and density of the air in the region between the shock wave and the cone. These quantities were evaluated for different values of the cone angle for a typical case. From the same theory the ratio of the yaw of the shock wave to the yaw of the projectile was determined, and was found to be in excellent agreement with values measured on spark photographs of moving projectiles.

Assuming that the theory would apply with reasonable exactness to an actual conical-headed projectile in flight in the neighborhood of the conical head, the normal head-force coefficient was computed for a series of values of the velocity and for cones of three different angles.

- (1102) *Petrología estructural: un método nuevo de geología*. Earl Ingerson. *Bol. soc. geol. mexicana*, vol. 13, pp. 41-60 (1948).

This is the first account in Spanish of the tenets, techniques, accomplishments, and potentialities of structural petrology. It discusses such fundamental concepts as the production of preferred orientation of mineral grains in rocks by growth and by differential motion, and defines special terms, such as "tectonite," "atectonite," and "fabric axes." The techniques of petrofabric analysis are described and illustrated.

Applications of the methods of structural petrology to problems of igneous, sedimentary, and metamorphic geology and to problems of economic significance are outlined from the literature in sufficient detail to show what the problems were and how these methods were able to contribute to their solution data not obtainable by other known techniques.

- (1103) A petrographic criterion for the possible replacement origin of rocks. F. Chayes. *Amer. Jour. Sci.*, vol. 246, pp. 413-425 (1948).

In rocks formed by replacement the amounts of residual and introduced materials

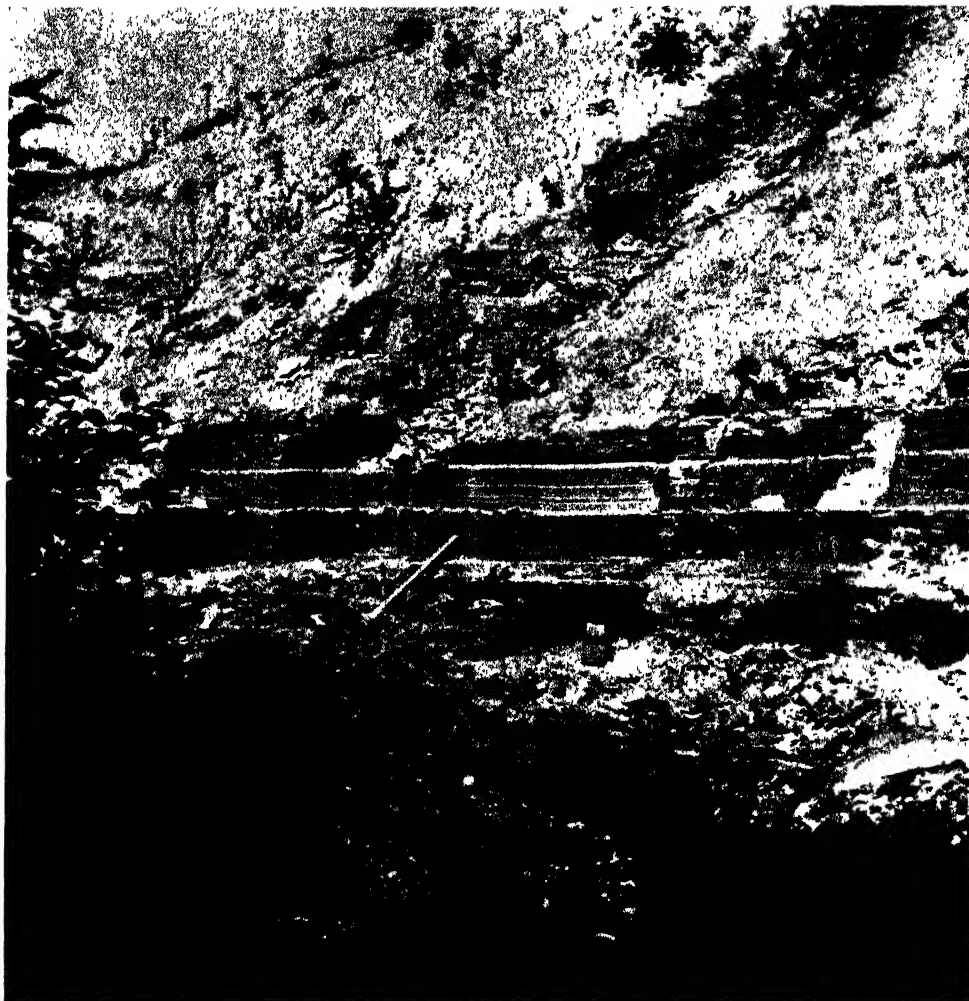
ought to vary inversely. This implies negative correlation between the two types of material, and it is argued that unless the negative correlation is significant, the sample from which it is computed may not be considered compatible with the replacement hypothesis in question. On this basis it is shown that varia-

tions in the chemical composition of the rapakivi granites are incompatible with Backlund's suggestion that these rocks have been formed by reaction between the Jotnian sandstone and alkali-aluminate emanations.

(1108) Annual report for 1947-1948.

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Glacial varves exposed in brook ravine at South Newbury, Vermont, by DTM magnetic expedition of 1947. Layers shown were deposited about 11,000 years ago, when glacial lake covered the Connecticut River Valley.

DEPARTMENT OF TERRESTRIAL MAGNETISM

Washington, District of Columbia

MERLE A. TUVE, *Director*

INTRODUCTION

With the completion of many volumes of survey and observatory results, and the transfer of the observatories to other agencies, the Department during this report year has begun to function effectively as a physics department with special research interests in geophysics and in laboratory physics. A program characterized by this view was chosen as best suited to the postwar situation and research interests of the individual staff members, and recognizes the responsibility which goes with the freedom of an endowed institution in the world of research today. A deliberate aim to work as physicists, in reasonably fresh areas, on significant problems, however difficult, and on problems directed toward philosophical goals without reference to possible applications, is the general policy adopted for guiding the selection and emphasis of the work carried forward. This is a high standard, but it is in accord with our belief that genuinely

creative research is always a spontaneous activity of an individual whose intellectual interest has been challenged by a problem. He can be encouraged, and his efforts can be greatly enhanced by the regenerative interest of his colleagues, but the primary motivation of a research man is the intellectual and aesthetic satisfaction he derives from personal participation, on his own initiative, in the problem which has challenged his interest and professional capacities. This policy gives rise at once to a highly varied program, but the close interplay in a small group, dependent on one another for technical help and facilities, has already led in a natural way to a concentration of interest by groups among the staff members of the Department, and to marked vigor and personal interest in each thrust toward a difficult problem. The report year ends with a marked degree of fresh initiative and enthusiasm.

SUMMARY FOR THE REPORT YEAR 1947-1948

Direct evidence for a mechanism by which charged particles may acquire energies in the cosmic-ray spectrum was provided by the discovery in 1946 at the Department of a few striking increases in cosmic-ray intensity during intense chromospheric eruptions of long duration. Study of changes with time in the magnetic fields of sunspots, from Mount Wilson Observatory data, indicates that these changes are capable of accelerating particles to energies in the cosmic-ray spectrum. This view was strengthened by theoretical investigation of the mechanism

by which the accelerated particles might escape from the sun in spite of the blocking effect of the sun's general magnetic field. This first investigation showed that the magnetic moments of a sunspot pair could, if properly oriented, create a tunnel into the region through which the passage of such particles would normally be forbidden. In this investigation the rare combination of the necessary accelerating voltage and the tunnel existed only at the times of the unusual increases in cosmic-ray intensity which were observed, and these requirements were not met

when certain other solar flares occurred which showed no cosmic-ray effects.

The seismic program, undertaken in co-operation with the Geophysical Laboratory, the U. S. Navy, and the New Mexico School of Mines, for investigating the structure and strength of the earth's crust has progressed to a point where suitable instruments have been developed and extensive field observations have been made in the region around Washington, D. C., and the Appalachian Highlands. About 140 satisfactory records have already been obtained in this area at distances up to 350 km from detonation of TNT charges of from 600 to 4000 pounds, and the evidence shows that distances certainly up to 1000 km and probably up to 1500 km are within the range of the apparatus when used in connection with large blasting operations.

The results indicate that the granitic layer in the region investigated is about 10 km thick and that the compressional wave velocity in it is between 6.0 and 6.17 km/sec. This is somewhat higher than the velocity usually assigned to granite in earthquake studies. The depth to the Mohorovicic discontinuity here is indicated as approximately 42 km, below which the velocity is 8.15 km/sec. Between the granite and the 42-km depth at least two intermediate basaltic layers are noted.

In addition to the refraction records, vertical and critical-angle reflection records have been obtained from a number of quarry blasts in the Shenandoah Valley. So far as the grosser structure is concerned, the reflection records to date appear to corroborate the conclusions reached from the refraction data.

Preliminary results from a much less extensive series of recordings in the vicinity of Albuquerque, New Mexico, indicate a greater thickness, around 14 km, and a

lower velocity, 5.4 km/sec, for the granitic layer in that region.

Electric surveys over the tops of thunderheads were begun for the purpose of ascertaining whether thunderstorms supply electricity to the earth at the rate required to maintain the negative charge, and its counterpart, the electric field, which is observed at the surface in all areas where fair weather prevails. A B-29 assigned to this project by the Air Weather Service of the U. S. Army Air Force was equipped with apparatus for measuring the following elements: (a) the electrical conductivity of the air, (b) the electric current flowing to an insulated plate on a wing, and (c) the vertical component of the electric field strength.

The altitudes at which the tops of the thunderheads were scanned during the first four flights, in October 1947, ranged from nearly 40,000 feet on the first flight to 44,000 on the last. These flights have demonstrated the feasibility of the project in most respects. The survey of the most active thunderhead at one stage indicated a maximum current of the right sign and of about the magnitude required for the theory that thunderstorms supply the negative charge on the earth. Improvements in the instruments were made during the winter, and the techniques and procedures have been made ready for more extensive observations during the summer and autumn of 1948.

In connection with the investigation of the magnetization of glacial clays, it has been found that the polarization has remained constant in direction and intensity since the clays were deposited, 10,000 to 20,000 years ago. This conclusion is confirmed by laboratory experiments in re-depositing the clays and testing their stability as well as their high coercive force. The consistency of the results for ocean-bottom sediments makes it prob-

able, or at least possible, that these sediments may also have remained constant in polarization since they were deposited.

It may be tentatively concluded that the horizontal direction of the earth's magnetic field has remained substantially constant during the past million years, and that the intensity has also remained substantially constant, allowing for a rather large experimental error, during the past 15,000 years. Work is in progress to extend the period of measurement to times of the order of one hundred million years and to determine more accurately, by laboratory investigations, the variation in intensity of the earth's magnetic field with time.

Significant progress is being made in improvement of the resolution of radio echo sounding methods in ionospheric research. The sweep speed of multifrequency techniques has been increased by a factor of 10, and the ultimate theoretical limit on speed of observation by pulse methods is being approached. These improvements will permit detailed study of the rapidly moving clouds of particles found by Wells and reported last year. With the success of this development, a three-year program has been planned for more detailed observation of these new effects in arctic and equatorial regions.

A new method of radio ionospheric recording is under investigation to simplify the present complex procedures of recording, reduction, and analysis of multifrequency records obtained during continuous recording. It now appears feasible to "lock on and follow" any dominant ionospheric characteristic, and record the results by a simple pen-and-ink recorder. The development of this type of recorder promises to have a substantial influence

on the study of the physics of the outer atmosphere.

Techniques are also under development for measuring the earth's magnetic field at high altitudes. Work is proceeding on both radio and rocket methods. Preliminary successful rocket shots have already been made in collaboration with the Applied Physics Laboratory of the Johns Hopkins University.

Progress has been made on further studies of the nuclei of light elements. Measurements on the angular distribution of the alpha particles produced in the disintegration of lithium have been extended to 3.5 million volts, using the large electrostatic generator. The results are in fair agreement with theoretical curves.

The biophysical program has continued its evolution to the point where the framework of an unusual research group has been developed. Gradual establishment of a program of studies of the fundamental properties of living matter is in progress. For the present a considerable number of projects employing radioactive tracers are under way involving some fifteen collaborators in the Washington area.

Among the achievements of the report year is the publication of the huge amount of observational material accumulated during the past quarter-century. The publication program set up in 1946 has now been essentially completed. At the close of the report year, two special treatises on the earth's magnetic field and ten large volumes of the *Researches of the Department* had been published, and an additional volume was scheduled to appear before the end of 1948. The greater accessibility of these data, so fundamental to analytical and theoretical studies, should go far to encourage theoretical research in geophysics.

EXPERIMENTAL GEOPHYSICS

THE EARTH'S CRUST

PALEOMAGNETISM

In order to determine the origin and nature of the earth's magnetic field and to test the various hypotheses which have been advanced to explain this field, it is desirable to know its history throughout geologic time and to investigate more carefully its special variations, both inside and outside the earth's surface. A small group in the Department has been actively engaged on the determination of the history of the earth's field, based on studies of the present polarization of crustal material and its magnetic stability. Unconsolidated fresh-water sediments have been investigated. These sediments are in the form of clays and offer one of the simplest types of polarization, since the clays can be redeposited under laboratory conditions. A particularly lengthy investigation of polarization of glacial varves has been made, together with measurements on core samples of sediments from the Pacific. From a study of anomalous deposits in the glacial clays, the geologic stability of the polarization of these clays has been established over geologic time.

From the measurements of the glacial clays it is concluded that the earth's field has not changed substantially in direction or intensity during the past 15,000 years. From measurements of the Pacific cores it is tentatively concluded that these elements of the earth's magnetic field have probably remained substantially constant during the past million years. A much more complete investigation is required to verify these tentative conclusions. It would be desirable to extend the measurements over long epochs of geologic time. The results so far obtained are not inconsistent with the "fundamental" theory proposed

by Schuster, Babcock, and Blackett, but do not provide any evidence to support this theory.

Statement of the problem. The origin of the earth's magnetic field is not yet explained. It is difficult to obtain adequate experimental data for describing the field sufficiently to permit making a test of the various hypotheses regarding its nature and origin. Two general types of theories have been advanced to explain the magnetic field of the earth. One assumes that the origin of the field is fundamentally connected with the mass and rotation of the earth and may be an inherent property of all rotating matter. These theories, at best, cannot explain the asymmetrical features which are observed. The second general class of theories attempts to explain the origin of the earth's field on the basis of complex fluid motions and related phenomena within the earth. In order to test the validity of these basic hypotheses it would be extremely valuable to have more complete knowledge of the variation of the direction and intensity of the earth's magnetic field with time, and also of the variation of the field deep within the earth.

The purpose of the Department's current effort is to provide as many such data as possible. To be useful, the period for which the history of the field is to be determined should be of the order of geologic time. It would be highly desirable to have this history for at least 100 million years.

Much work has been done by previous investigators on the study of the magnetization of baked clays and of rocks; the literature contains hundreds of names and goes back more than 250 years. One of the most recent and comprehensive discussions of the subject, published in 1942 by Dr. Hans Haalck, emphasizes the

danger of drawing conclusions as to the direction and intensity of the earth's magnetic field in former times from measurements on individual samples, and our work confirms this view.

Description of apparatus. The method employed to measure the polarization of the clay samples is the same as that described by Johnson and McNish (Year Book No. 38 [1938-1939]). The sample is rotated close to the center of a coil, and, acting as a small magnetic dipole, induces an alternating voltage in the coil. The intensity and phase of the signal are measured, after amplification, and from these measurements the magnetization of the sample can be deduced. The sample, in the form of a cylinder 4 cm in diameter and 4 cm long, is rotated at the end of a shaft at 10 cycles per second.

The apparatus, except for the electronic equipment, was constructed of nonmagnetic materials and very ruggedly built to withstand vibration and shock during transport in a large laboratory truck which is taken to the sites where the clays and rocks are found. The sensitivity permits the detection of a moment of 2×10^{-8} cgs unit per cubic centimeter. For moments of 10^{-6} cgs unit per cubic centimeter, the direction of magnetization can be determined to within one degree.

The clay samples discussed were obtained from the glacial clays of New England at sites fully described by Antevs following his work in 1922-1926. He identified about 5000 layers or varves in the Connecticut Valley region and assigned to the earliest layer, which he located near Hartford, Connecticut, the arbitrary date 3000. The latest varves were located in Vermont and dated about 8000. From recent radioactivity studies of these clays by W. D. Urry, of the Geophysical Laboratory, the arbitrary dates 3000 and 8000 have been found to correspond to actual

dates 15,000 and 10,000 years, respectively, before the present epoch.

To begin the sampling, the horizontal surface of a varve was exposed; in most cases this was quite simple, since the varves would separate readily at a sandy boundary. Many of the layers thus exposed were not truly horizontal, but had so-called "tilts" up to several degrees. The word "tilt" has been adopted here to describe the departure from the horizontal which geologists call "dip," because in magnetic studies the word "dip" commonly refers to the inclination of the earth's magnetic field at any point.

Method of magnetization of clays. It is supposed that the sedimentary clays obtain their original polarization in the following way: The sediments in general contain only a very minute amount of magnetic material. In the case of the clays deposited in the glacial lakes of New England, the magnetic material is principally magnetite. As these minute particles settle in still water, they are subjected to gravitational, thermal, hydrodynamic, and magnetic forces. The magnetic forces tend to align the particles in the direction of the earth's magnetic field. Thermal forces tend to produce a random orientation of the particles and prevent any polarization of the clay. The gravitational and hydrodynamic forces tend to alter the direction of the particles, depending on their shape, away from an exact orientation in the direction of the existing field of the earth, but particles of an appropriate range of sizes will retain an average orientation after settling on the bottom.

Laboratory deposition of clays. The laboratory investigation of glacial clays has been carried out by using a very thin suspension of clay in water, and allowing it to deposit in a controlled magnetic field within a nonmagnetic container.

The results of the measurements on these samples were as follows: (a) The horizontal direction of polarization of the laboratory clays was always exactly the direction of the present horizontal magnetic field within the experimental error of measurement. (b) The angle between the horizontal and the magnetic vector of the polarization was always somewhat less than the angle of dip of the applied field. This difference was as much as 20° for weak fields, but decreased progressively as the field strength increased. (c) The magnetic polarization was a definite and reproducible function of the applied magnetic field. (d) Measurements made at several different water temperatures indicated a modest change of polarization with temperature. Experimental difficulties in measurement have so far prevented an accurate determination of this temperature coefficient, which, in any event, is small. From these measurements it is concluded that the initial polarization of the clays reproduces faithfully the horizontal direction, but only imperfectly the dip of the ambient magnetic field in which they are laid down.

Samples of both natural and artificial varves were shaken on a shaking table with amplitudes equivalent to those of very strong earthquakes over periods of 12 hours—thus simulating the effects of many centuries of earthquakes in the New England region. The samples were placed in various possible orientations, including that at right angles to the ambient field of the earth. The samples tested were both dry and very wet. No change in polarization within the experimental error was observed. These laboratory experiments indicate very considerable stability of polarization of the clays.

Intensity. One determination of intensity of the earth's field at the time of deposition of the glacial clays has been made. The

clay used was collected at South Newbury, Vermont (see plate 1), and represented deposition over a period of 200 years. The polarization was measured and an average value for these years was obtained. Clay from the same years was thoroughly mixed with water and redeposited in the laboratory under varying field strengths. The value for the ambient field corresponding to the natural polarization of this clay is the same as that for the earth's field at the present time, within the experimental error of measurement. The age of this clay, according to a preliminary determination by W. D. Urry, is 10,000 years. Antevs estimated its age to be about 27,000 years.

According to this measurement the intensity of the earth's magnetic field has not changed more than 0.5 per cent per century, a figure which differs markedly from the 5 per cent per century found from measurements at magnetic observatories during the past two or three hundred years. The intensity of magnetization of the New England clays varied, depending on locality, about twenty-fold. The maximum intensity observed was 40×10^{-5} cgs unit per cubic centimeter, and the minimum observed was 2×10^{-5} cgs unit per cubic centimeter.

Stability. Stability of the polarization of the glacial varves at South Newbury was investigated in the following way. Between the layers representing the years 7006 and 7008 in Antevs' chronology, an anomalous deposit of clay was found in excavating. This material consisted of varved clay which had been mixed and kneaded and then deposited with the layers randomly oriented. This abnormal material existed in patches. Since the layers below and above these patches coincided exactly with the normal series existing in near-by exposures, it is firmly established that this clay was deposited in some special way at that time, and

before the next layers were deposited. Thus the disturbance in the layer occurred at the time of the deposit, and any magnetic irregularities in this disturbed material as compared with the normal layers have persisted over the entire period since the time of deposit.

If the polarization in the normal layers is unstable and has changed so as to follow the ambient field of the earth in direction and intensity, then the polarization of the disturbed layer should, during the past 10,000 years, have also been magnetized substantially in the same direction and with the same intensity as the normal layers. On the other hand, if it has retained a more or less statistical magnetization, as would be indicated by its stirred and mixed nature, then the magnetic direction and intensity of this clay should be much more nearly statistical. The intensity, for example, would be expected to go from nearly zero to approximately the same value as that of the normal layers, and the magnetic direction should show great local variations among samples. The measurements show conclusively that various parts of the disturbed layer have retained over geologic time a substantially random polarization as compared with the normal layer. It is thus concluded that the various layers of glacial clays at South Newbury have retained essentially the polarization which they obtained when they originally settled in the glacial lake.

Declination. The results for declination cover a period of about 5000 years. One thousand measurements are included. Over this period, the data show that the declination remained substantially constant on the average, with excursions of the order of $\pm 30^\circ$. These excursions are consistent with the measurements made at London, Boston, and Baltimore during the past few hundred years. The average declination over the 5000 years is $\pm 10^\circ$

from the present declination, which is about -15° in New England. There is some slight suggestion of a periodicity of the order of about 300 years.

Inclination. As yet there is little confidence that the values of inclination measured in the glacial varves have an exact relation to the inclination at the time of deposition. These doubts arise largely from the results of the laboratory settling of the clays.

Measurements on Pacific Ocean cores. During 1947, six Pacific Ocean bottom cores were received through the courtesy of the U. S. Navy Hydrographic Office from among those obtained on the U. S. Antarctic Expedition of 1946-1947. The length of the cores ranged from 150 to 250 cm. The magnetic characteristics of five of these were studied, one of which was obtained near the Galapagos Islands, one near Easter Island, and the remaining three below the Antarctic Circle near the international date line. The depth at which the cores were taken ranged from 1635 to 2150 fathoms.

Successive cubical samples were cut throughout the length of each core, 400 samples being obtained from the five cores. The sediments in the cores were dated by radioactive equilibrium measurements by W. D. Urry, of the Geophysical Laboratory, and one core was found to have only 170,000 years of deposition; three represented about one million years, and the remaining one fell between these limits. In none of the cores did measurements of magnetic declination differ markedly from present values, the assumption being made that the top of each core had the present declination for its particular locality. These results are tentative because some aspects of the results call for further interpretation. Inclination or "dip" measurements on a limited number of the cubical samples from each core have indi-

cated conformity with present values for this element for each core location. Thus, for the core from near the Galapagos Islands, the inclination of the field from the horizontal plane (negative because the south pole dips downward) is small (-20° or less), whereas for the Antarctic Circle cores the field is nearly vertical (-60° to -80°). The measurements on the cores thus have indicated that the earth's magnetic field has been much the same as it is now for the past million years. In the absence of proof of the magnetic stability of these cores, this result cannot be accepted as an established fact, but the interlocked character of the magnetic particles in the sediments, together with the stability indicated by the folded varves, gives this interpretation considerable weight. All the ocean cores were weakly polarized as compared with the New England clays, but the high sensitivity of the measuring apparatus made satisfactory measurements possible.

SEISMIC SOUNDING

Prewar investigations and plans were broken up by so many years of national defense activities that most of the staff members began their postwar plans with a fresh outlook, relatively detached from their previous activities. In addition, there became available a considerable amount of war surplus material, some of which made it possible to initiate long-desired research programs which had previously been out of reach.

One such program was a more intensive and thorough investigation of the structure and strength of the earth's crust under the different kinds of loads which exist in different geographical regions. Particular problems are the kind or kinds of compensation underlying mountain regions, the geological nature of large gravity

anomalies, and the correlation of isostatic adjustment with crustal layering. The method proposed for an approach to these questions was to use the best combination of refraction and vertical and critical-angle reflections of seismic waves resulting from controlled detonations of high explosives.

The Geophysical Laboratory and the Department of Terrestrial Magnetism determined to initiate such a program as part of their experimental studies of geophysics of the earth's crust. The Carnegie Institution thereupon requested the U. S. Navy to earmark a sufficient quantity of high explosives for the purpose. The Navy not only complied with this request, but also, through several of its branches, has collaborated in firing the charges, in procuring certain equipment, and in work on instrumentation problems. Other collaborators are the New Mexico School of Mines (E. J. Workman) and Columbia University (M. Ewing). It is hoped that many other groups will participate in these studies.

The first phase of the investigation was concerned with the development and field testing of suitable instrumentation and techniques. This is now completed except for possible refinements.

The second phase involved a planned series of field observations of seismic waves from detonation of tamped high explosives. The first objective was to determine the layering of the crustal structure underlying the region around Washington, D. C., and the Appalachian Highlands. The explosive charges fired for these tests ranged between 600 and 4000 pounds, and 140 "good" records, which are listed in tables 1 and 2, have been obtained out to 350 km distance. In the three columns under "first arrival" (and similarly for second and third arrivals) are listed times of travel, apparent velocities, and amplitudes of ground motion in

TABLE 1
SUMMARY OF SEISMIC RECORDS TO MARCH 31, 1948

LBS. EXPLOSIVE	FIRING POINT	SEISMOMETER STATION	Δ HORIZ. DIST. (km)	No. RECORDS	BACK- GROUND MOTION (microcm)	FIRST ARRIVAL			SECOND ARRIVAL			THIRD ARRIVAL		
						$T-T_0$ (sec)	$\Delta/T-T_0$ (km/sec)	Ampl. (microcm)	$T-T_0$ (sec)	$\Delta/T-T_0$ (km/sec)	Ampl. (microcm)	$T-T_0$ (sec)	$\Delta/T-T_0$ (km/sec)	Ampl. (microcm)
60	Mt. Weather, Va.	Mt. Weather, Va.	2.6	1	.	(0.4+)
60	Mt. Weather, Va.	Mt. Weather, Va.	5.1	1	.	0.83	6.16
2800	Perry Quarry, Va.	Along strike	6.76	1	.	1.14	5.93
		Across strike	14.47	1	.	2.56	5.64
		Along strike	16.84	1	.	2.71	6.22
		Across strike	31.72	1	.	5.20	6.10
2000	Albuquerque, N. M.	Albuquerque, N. M.	22.6	1	.	3.96	5.72
1200	Patuxent River, Md.	Cheltenham, Md.	52.9	3	.	9.7	5.46
600	Solomons, Md.	Eastern Shore, Md.	61.4	8	.	10.9	5.63 \pm 0.05
	Atlantic, off Ocean City, Md.	Washington, D. C., DTM	84.1	13	.	14.0	6.0	...	16.4	17.1
1200	Patuxent River, Md.	Washington, D. C., DTM	85.5	2	.	14.3	5.98
4000	Chesapeake Bay, off Little Cove Point, Md.	Washington, D. C., DTM	92.0	3	.	15.1	6.1	...	17.1	(18.2)
4000	Chesapeake Bay, off Cove Point, Md.	Derwood, Md.	102.3	21	0.35	16.9	6.05	0.22	18.6	...	1+
600-2400	Patuxent River, Md.	Englewood, N. J. (Ewing)	125.0	1	.	20.7	6.05
32,000	Rock Hill Quarry, Pa.	Gambrell Park, Md.	150.7	4	0.21	24.8	6.07	0.6
600	Patuxent River, Md.	Hagerstown, Md.	181.1	7	0.13-0.6	29.0	(6.25)	0.7	(29.9)	6.05	1
600-1300	Patuxent River, Md.	Chambersburg, Pa.	201.1	2	...	31.6	(33.1)	6.07
1200	Patuxent River, Md.	Mercersburg, Pa.	205.0	4	0.07	31.6	...	0.7	33.1	6.18
600-1200	Patuxent River, Md.	Carderock, Md.	228.3	1	.	36.9	6.18
32,000	Rock Hill Quarry, Pa.	Derwood, Md.	235.0	14	0.15-0.3	38.7	6.04 \pm 0.07	0.3-0.45
600	Atlantic, off Ocean City, Md.	Shade Gap, Pa.	236.0	8	0.07-0.25	36.15	(6.53)	0.13	37.4	...	0.04	(39.1)	6.04	0.03
1300-2400	Patuxent River, Md.	Loyburg, Pa.	259.0	2	0.04	39.0	(6.66)	0.02	39.6	6.55	0.025	42.2	6.14	0.1
2400	Patuxent River, Md.	Blue Knob, Pa.	275.7	2	0.02	42.3	(6.52)	0.01	44.9 \pm	6.15	0.025
2400	Patuxent River, Md.	Laurel Hill Park, Pa.	300.1	5	0.02	45.3	(6.62)	0.01	46.1	(6.5)	...	(48.5)	6.2 \pm 0.05	0.02

TABLE 2
SUMMARY OF SEISMIC RECORDS APRIL AND MAY 1948

LBS. EXPLOSIVE	FIRING POINT	SEISMOMETER STATION	Δ HORIZ. DIST. (km)	No. RECORDS	BACK- GROUND MOTION (microcm)	FIRST ARRIVAL			SECOND ARRIVAL			LATER PHASES
						T - T ₀ (sec)	Δ/T - T ₀ (km/sec)	Ampl. (microcm)	T - T ₀ (sec)	Δ/T - T ₀ (km/sec)	Ampl. (microcm)	
1200...	Patuxent River, Solomons, Md.	Cheltenham, Md.	52.9	2	0.6	9.4	5.62	1.0
1200...	Patuxent River, Solomons, Md.	Washington, D. C., DTM	84.1	3	0.7	14.0	6.00	1.0	21.6 (ampl. 3.6)
1200...	Patuxent River, Solomons, Md.	Widewater, Md.	94.6	3	0.01	15.8	6.00	0.05	17.5	0.5	20, 21.4, 22.5
1200...	Patuxent River, Solomons, Md.	Seneca Creek, Md.	109.7	3	0.1	18.1	6.06	0.2	19.3	0.3	20.2 (ampl. 1.5), 21.4
1200...	Patuxent River, Solomons, Md.	Boyd's, Md.	114.9	3	0.03	19.1	6.03	0.6	20.2	0.6	21.1, 26.5
1200...	Patuxent River, Solomons, Md.	Kingsley, Md.	121.7	3	0.01	20.3	6.01	0.4	21.1	1.0	22.1 (ampl. 1.8), 23.5
1200...	Patuxent River, Solomons, Md.	Sugarloaf Mt., Md.	128.0	3	0.15	21.1	6.06	1.5	25.0	2.0	27+, 29
2400...	Patuxent River, Solomons, Md.	Pine County, Pa. (near Reading)	239.3	2	0.1	38.5	6.22	0.2	40.8	0.6
2400...	Patuxent River, Solomons, Md.	Chestnut Ridge, Pa. (near Ligonier)	322.1	2	0.05	51.6	6.25	0.1	53.4	0.15	55.1 (ampl. 0.2), 78
2400...	Patuxent River, Solomons, Md.	Lake Mohawk, Sparta, N. J.	335.2	2	0.06	49.2	(6.82)	0.06	55.4	6.20	0.1
2400...	Patuxent River, Solomons, Md.	Elliot State Park, Pa.	354.3	2	0.07	51.0	(6.94)	0.06	57.1	6.21	0.15	55.8, 60

microcentimeters (excluding frequencies below 1 cycle per second).

The amplitudes observed depend not only on weight of charge and distance, but also on the degree of coupling of explosive charge and seismometer with solid bedrock. In addition, apparent abnormalities are observed at certain distances owing to the simultaneous appearance of several phases. In general the

tables 1 and 2 and are shown in figure 1. The seismic velocities in the various layers and the depths of the discontinuities may be obtained from the slopes of the curves and the distances at which they intersect. The subcrustal layers and compressional wave velocities in the region around Washington, D. C., appear to be as follows: 0-10 km depth, velocity 6.0-6.17 km/sec; 10-24 km, 6.7 km/sec; 24-42 km, 7.05 km/

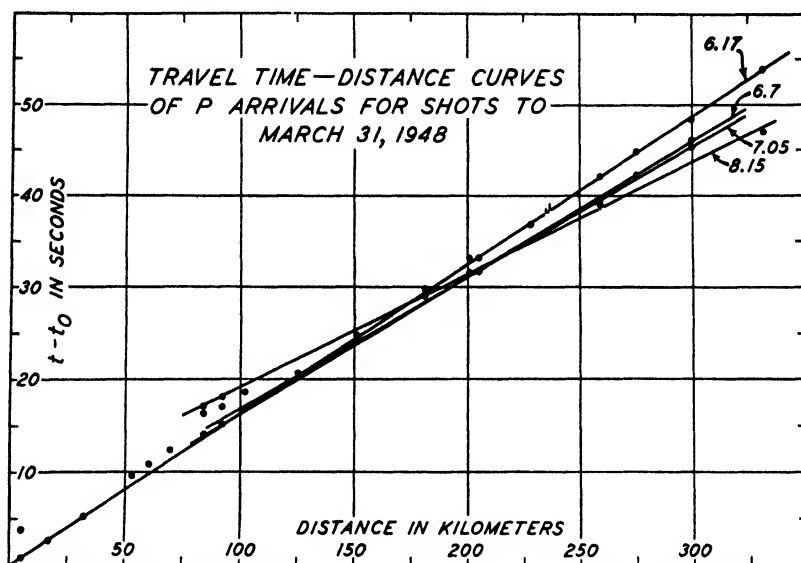


FIG. 1

amplitudes of first arrivals between 100 and 400 km distance lie between 10^{-6} and 10^{-8} cm. The apparent predominant frequencies lie between 4 and 15 cps, but higher frequency components are present which are essential to good recordings. Under very favorable circumstances, which require avoidance of man-made and wind disturbances, seismic amplitudes down to almost 10^{-8} cm at the greater distances can be observed and utilized. This amplitude is less than a thousandth of a wave length of visible light.

Curves showing travel time versus distance have been drawn from the data of

sec; below 42 km, 8.15 km/sec. This layering is shown graphically in figure 2, which also shows some of the seismic wave paths as dashed lines.

From classic plane wave theory the partitioning of energy, and thus of amplitude, of refracted and reflected seismic waves may be computed if the density and velocity relations across the discontinuity are known. Although not rigorously applicable, these computations provide a very valuable semiquantitative basis for intelligent planning of experimental field work for recognition of later reflection arrivals. In figure 3, after Knott (1899),

are plotted the reflection coefficients as a function of horizontal distance for a certain assumed vertical discontinuity in velocity between layers of the crust. From such considerations, assuming the feasi-

indicated above regarding the grosser aspects appear, however, to be corroborated by observations of strong reflections at appropriate critical angles. This investigation of reflection observations, and

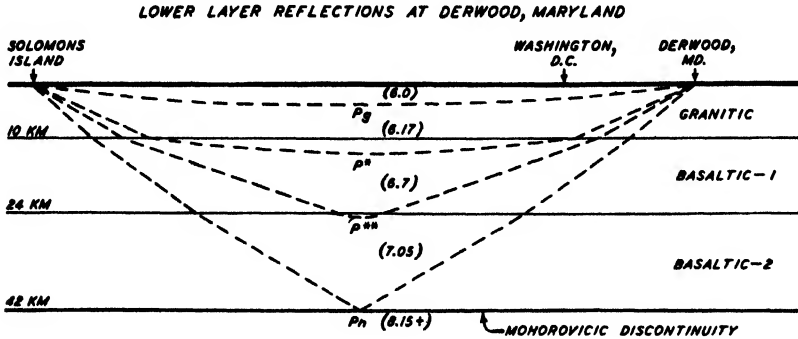


FIG. 2 .

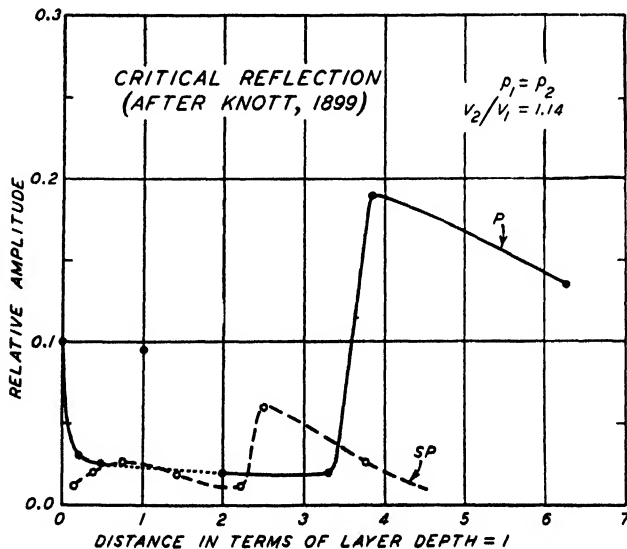


FIG. 3

bility of correct interpretations, the special value of reflection observations for "point by point" surveys should be readily apparent.

Results from reflection shooting are more conflicting because they are able to indicate finer details of structure and are, therefore, more complex. The conclusions

in particular of vertical reflections, is being vigorously pursued. For this purpose, amplifiers have been modified to operate at the high gains required for later phases and yet permit large overloads from first arrivals without damage to the recording pens.

No intensive investigations have yet

been made of shear and surface waves. A preliminary survey indicates, however, that these are of much longer period and have velocities approximating those calculated on the assumption of 0.25 for Poisson's ratio.

With the co-operation of the New Mexico School of Mines (Research and Development Division), seismic observations were made in the vicinity of Albuquerque, New Mexico. Energy losses from the explosions were high, but data were obtained up to 55 km from the shot point. From the data obtained, it was found that the top layer in the region of the Manzano Mountains was 14.4 km in depth and the velocity for this layer was approximately 5.4 km/sec. At the time of this report the New Mexico program is still in progress, and the results thus far obtained have not been completely analyzed.

THE EARTH'S ATMOSPHERE

THE EARTH'S ELECTRIC FIELD

The Electric Field above Thunderheads

The chief purpose of this project is to ascertain whether thunderstorms supply negative electricity to the earth at a rate sufficient to maintain the negative surface charge and the corresponding electric field intensity that have been found to prevail in all fair-weather areas.

C. T. R. Wilson was the first to suggest that thunderstorms might be the source of the supply current, and F. J. W. Whipple adduced some qualitative evidence which gives a measure of support to that suggestion. The question "Is the net electric current from a typical thunderstorm to earth of the right sign and magnitude?" has not been definitely answered. The interesting attempt made by Wormell to draw up a "balance sheet" for the various processes of electric interchange

between air and earth under a thunderstorm was designed to give an answer to that question. The affirmative answer which was indicated is, however, based on several categories of data, some of which may not be representative of the typical or average thunderstorm. An overwhelming complex of circumstances would be encountered in any attempt to make, beneath a thunderstorm, a survey of the net transfer of electricity from air to earth. In fact, such an attempt has apparently never been made. It would require the measurement of a number of elements, some under unfavorable circumstances, at each station of a network extensive enough to ensure that the full set of measurements would give a reliable cross section of storms which traverse the network. Such cross sections for a number of storms would be required.

Another scheme for finding the answer to the question posed depends on the fact that the net vertical current well above the thunderhead must be of the same sign and of the same average magnitude as that below. The electrical conditions above the thunderhead are also simpler than those beneath it. Lightning seldom, if ever, strikes from the top upward, and, of course, precipitation does not participate in transfer of electric charge there. All that is required is a survey of the electric conduction current. The realization of this scheme requires suitable electric equipment and an airplane that can be operated at altitudes of 40,000 to 50,000 feet (12 to 15 km) or higher. Thus equipped, it is unnecessary to wait passively for a thunderstorm to come up, since any storm that is within a radius of several hundred miles of the air base may be overtaken and surveyed.

Such was the outlook when the project was proposed in November 1946. Promise of co-operation of the U. S. Army Air

Force in the project was received early in 1947. The necessary instruments were developed and tested in the laboratory, and late in September 1947 a test flight was made in a B-29 with all the equipment installed. The plane was based at Tinker Field near Oklahoma City, Oklahoma, toward the end of the thunderstorm season, and during October 1947 four flights over thunderheads were made. On these flights, surveys lasting an hour or more were made over thunderheads at altitudes ranging between 40,000 and 44,000 feet. Though these are not altitude records, the length of sustained flight and the total time of flying at these altitudes for one crew and one plane are decidedly unusual.

The equipment was designed to measure and register elements from which the vertical component of the electric conduction current could be derived. The total current flowing out of the top of the thunderhead was then derived by a rough integration of all the values measured in the traverses back and forth from various directions over the center of the thunderhead.

The instruments were as follows: (a) two essentially identical apparatus for measuring the electrical conductivity of air, one for the part contributed by positive ions, and the other for the contribution by negative ions; (b) one device designed to give a direct measure of the conduction current; (c) two electric field meters kindly lent by the Naval Research Laboratory. These field meters were mounted on the right wing about one chord length from the tip, one above the wing and the other directly opposite below the wing. The true vertical field strength is obtained approximately by taking half the algebraic difference of the registrations for top and bottom exposures respectively after reducing these to a common basis.

The tubes in which the air is sampled for conductivity are situated in a streamlined structure attached to the underside of the left wing about one chord length from the tip. An insulated plate 1 foot in diameter, mounted under the right wing inboard from the bottom field meter, is used for collecting electric current "directly." In these five installations small electric currents, oscillating in the case of the field meters, are amplified in order to actuate microammeters located in the fuselage and arranged on a panel. This panel is photographed at regular intervals on 35-mm film.

The initial results are encouraging. The altitudes reached and maintained for the required time exceeded expectation. More maintenance was required than in the case of normal flight, but with the excellent maintenance service provided by the U.S.A.A.F. no conspicuous handicap was encountered. The pilot, Lieutenant Paul Crumley, and his crew deserve credit for this performance. The demands on the skill and judgment of the pilot were out of the ordinary, and the task was arduous and extremely hazardous. The design of equipment and the procedure for recording the prescribed elements were found satisfactory. A few mechanical or electrical faults which intervened at intervals during these flights have called for minor changes during the past winter in preparation for flights planned for the summer of 1948. The records, although affected in places by such faults, are generally acceptable.

Some deductions from the 1947 records which may be stated at this stage of the project are as follows: (1) The electrical conductivity of air increased with altitude up to 40,000 feet in a way consistent with that observed on the flight of the stratosphere balloon *Explorer II* (Year Book No. 37, 1937-1938). This is further evi-

dence that no correction of these measurements for pressure or temperature effects in the instruments is required. (2) The electrical conductivity of air over the thunderheads surveyed seems to be normal considering the altitude, although it averages somewhat lower than the *Explorer* values. (3) The electric current in the air over the thunderheads was relatively insignificant on most traverses. The traverse which showed the largest current density indicated a maximum current upward of about 1 ampere for the whole thunderhead. This is of the right sign and about the right magnitude to account for the negative charge on the earth if, as estimated by Brooks, 1800 thunderstorms are, on the average, in progress on the earth at a given instant.

During the 1948 season, with somewhat improved instruments and with the plane stripped of armament and equipped with new engines, it is expected to make surveys during thunderstorms over Ohio within range of the radar V-beam network at Clinton County Air Base.

Atmospheric Conductivity in Peru

During July and August 1947, continuous recordings of small, intermediate, and large ion densities, and rate of ion formation, were made at the Geophysical Institute of Huancayo, Peru. Atmospheric conductivity and potential-gradient recorders, previously discontinued, were also run during this period. A complete analysis of the data thus obtained has not yet been made, but some interesting points have arisen from the preliminary analysis.

From simultaneous recordings of small ion densities and conductivities (positive and negative), small ion mobilities were computed, and the ratio of values found to those generally accepted at sea level agrees with the inverse pressure law of mobility.

There is strong evidence for a diurnal variation in the mobility of ions of one sign (probably negative).

It appears likely that the anomalous diurnal variation in atmospheric conductivity can be completely explained by variations in intermediate and large ion densities, acting in accordance with the usual formula for small ion balance in the atmosphere. This pushes the problem back one stage without solving it, for we must now determine the origin of variations in large and intermediate ion densities. This may be difficult, for several types of ions with different mobilities appear to be involved.

Rate of ion formation in the atmosphere was found to be high, averaging about 40 ion pairs/cc/sec. The increase in ion formation during thunderstorms noticed by Wait and McNish in Washington was also observed, and the rate of decay of this extra source of ions confirmed their contention that it was due to radium B.

UPPER ATMOSPHERIC RESEARCH

Studies of the Upper Atmosphere

The atmosphere of the earth serves as an insulating envelope containing the earth, and shields its surface from much of the active corpuscular and electromagnetic radiation incident from outer space. In fact, of the electromagnetic radiations, only the one octave of light waves and some lower-frequency radio waves can penetrate this atmospheric envelope; of the corpuscular radiations, only the energetic products of cosmic-ray encounters with atmospheric elements succeed in reaching the earth's surface.

In insulating these active radiations from its surface, the atmosphere plays two important roles. The atmosphere contains the "earmarks" of their presence for the geophysicist to read with refined methods

of observation and interpretation; and as a consequence of absorption of these radiations secondary effects are produced in the atmosphere which lead to many other vitally important geophysical phenomena.

One of the more important tools for exploration of the outer atmosphere is echo sounding with radio waves. Since the introduction of this technique by this Department over twenty years ago and its subsequent development in multifrequency applications, the radio method has come into use for outer atmospheric observation in laboratories everywhere. Just as the simple microscope opened new worlds to the scientist, so has radio sounding given him a new opportunity for visualizing phenomena that connect us with outer space, processes which heretofore have been hidden by the opacity of the atmosphere. Moreover, this technique, in solving some of the earlier and more obvious problems, has, at the same time, raised a hundred new and more difficult ones which could not be foreseen. The development of radio sounding in the present forms, however, by no means exhausts the power of this tool for the exploration of the outer atmosphere.

In attacking these new problems it is possible to improve the resolution of the radio technique manifold, thus providing the necessary improved vision for the observation and interpretation of the more complex phenomena.

The postwar program of the Department of Terrestrial Magnetism in upper atmospheric research envisages a critical study of phenomena of the outer atmosphere which enter into the relations of the earth with the sun, the moon, and other elements of the universe. Since this study requires, as a first step, the development of new tools, the initial effort of this postwar era has been directed toward the

evolution of radio-sounding techniques for improved methods of observation.

Three problems of special significance have been selected. The first relates to the discovery by Wells and his colleagues of rapidly moving clouds of particles which seem to plunge into the atmosphere at times of magnetic disturbance. This discovery arose from an improved technique which speeded the sweep of the apparatus to twice each minute. It was immediately apparent that this speed was still too low for the proper resolution and delineation of the transient incidents accompanying disturbances. As a result of further effort during the past year, the speed of the apparatus has been increased by another order of magnitude, until the ultimate theoretical limit of resolving power of pulse-echo-type apparatus has been reached. This new research tool is at present being perfected, and a program for its application has been planned.

The second problem relates to the origin of magnetic disturbance in the atmosphere. In the endeavor to localize the source of disturbance, a means of measuring the level and magnitude of electric current flow in the outer atmosphere is necessary. This has led to investigation of radio methods for measuring the earth's field at great heights.

Closely allied to this research is the measurement of the earth's field at high altitudes by means of rockets. The two methods are complementary, for, while the former may be used to follow the variations with respect to time at a selected level, the latter can provide a relatively instantaneous cross section with respect to height, and the one method can be used to check the other.

The third problem relates to the need for a greatly simplified means of following continuously any selected dominant characteristic of the ionosphere. The basic idea

had its origin in the common interest of the Mount Wilson Observatory and the Department of Terrestrial Magnetism in the study of bright solar chromospheric eruptions.

Since the discovery by Forbush in 1946 of coincidence of cosmic-ray bursts with certain chromospheric eruptions, it has become especially desirable to know more of the special characteristics of such eruptions. It is now well known that unusual absorption in the ionosphere (radio fade-out) is a sensitive measure of solar chromospheric activity. It was proposed by the Department in 1937 that if a continuous index of absorption of the ionosphere could be maintained, it could provide immediate warning of chromospheric activity—a warning that would permit more adequate preparation for visual observation of the transient solar effects. This proposal has led to the concept of the duplex-channel differential recorder to provide such a warning, and as the development of this device has proceeded it has become evident that it will be capable of recording in a simple and direct way not only the absorption index of the ionosphere, but also many other selected dominant characteristics.

Panoramic recording of ionosphere. The formulation of a definite program for the panoramic recording of ionospheric characteristics has provided a means of exploitation of a new and powerful research tool, the fast-sweep recorder. Special provision has been made to permit planning over a period of several years both for completion of instruments and for subsequent field operations. The program may be resolved into the following phases: (1) completion of the prototype instrument; (2) construction of two panoramic recorders; (3) registration of detailed ionospheric structure at Washington; (4) operation at selected field stations in arctic

and equatorial regions; (5) analysis and presentation of results.

Substantial progress has been made toward the completion of a prototype recorder. Many of the problems encountered in development of the finished instrument fall into new and virgin fields of electronic applications. One example is the amplifier in the ionospheric transmitter, which has to develop several kilowatts of output power over a band width of 20 megacycles from a fraction of a watt input power. This has been accomplished without any mechanical devices such as band switches or tuned circuits. For the low-power stages, video amplifier techniques are adapted. For the high-power stages, the same basic principles are extended, but the problems have been new and more difficult.

The speed of recording has been stepped up by 600 per cent, from two to twelve observations per minute. Significant changes in the echo pattern continue to be observed, even at the 5-second speed. Provision is being made for pushing the recording speed to the theoretical limit for pulse equipment, which has been computed to be 3 seconds for the full (20-MC) band, and proportionally less if a smaller band is scanned. This maximum resolution is finitely established by parameters such as velocity of propagation of radio waves, maximum height and frequency ranges, and acceptable uncertainties in measurement. In order to achieve this ultimate resolution, it will be necessary to compensate for the change in frequency between pulse and echo, using proved techniques.

Material and components for some of the power supplies for the two new recorders are being assembled. Construction is scheduled to start in July 1948, and will be continued as soon as progress on the prototype permits. It is planned to make

recordings of ionospheric activity at the Derwood Experimental Laboratory during disturbed periods in order to obtain a fair sampling of mid-latitude characteristics. The present period of high solar activity is especially suitable for studies of rapid fluctuations.

Field observations are scheduled for parts of 1949 and 1950. The first installation is planned for one or two arctic sites, preferably in or near the zone of maximum auroral activity. Several months of operation will probably be required to obtain the desired data. An equatorial site is proposed for the next installation, probably at Huancayo, Peru. Data from these two extremes will be supplemented by intermittent observations at Derwood. They will be analyzed separately and collectively in a study of many fundamental processes of the ionosphere.

Observations of meteor trails in the upper atmosphere. Radio observations of meteor trails made as a spare-time investigation by a member of our technical staff, Charles A. Little, Jr., using a radar constructed by himself, show a distinct diurnal variation in the distribution of meteor occurrence with time of day. A variation of this sort has already been established not only by visual observations, but by radio observations of Appleton and Naismith and of Hey and Stewart. The seasonal change in the nature of this distribution as reported by these authors, however, is not confirmed, since the present observations, which were made between April 17 and June 19, 1948, correspond almost exactly to the published curves for midwinter rather than midsummer. Since the present observations are at a different latitude and at a later epoch than the earliest published observations, the seasonal distribution may be more complex than was originally supposed.

The new observations also show, sur-

prisingly, a diurnal variation in the height of the meteor trails. Whereas the height throughout the night hours centers around 75 miles and the scatter ranges within height limits of 65 to 90 miles, there is a sharp rise of the meteor trails to an average height of about 130 miles at 9 A.M. local time. Furthermore, at this time practically no meteors are seen below a height of about 100 miles. This variation has not been evident in any of the earlier work and suggests an important approach to the study of the physics of the ionosphere. Little's regular work is concerned with operation of the nuclear physics equipment.

Measurement of the Earth's Magnetic Field at High Altitudes

Radio methods. Three radio methods of measuring the earth's magnetic field at high altitudes have been proposed. All depend on the gyration of electrons under the influence of the earth's magnetic field.

(1) Measurement of separation of critical frequencies of ordinary and extraordinary components. This method can be used only in the vicinity of the geomagnetic equator because of the "sidewise skidding" of radio waves at levels of low ion gradient in latitudes where the inclination of the earth's field is appreciable.

(2) Direct measurement of the gyro frequency. This experiment depends on an effect of cross modulation between two radio frequencies in the ionosphere which was predicted in 1936 by V. A. Bailey. The experiment has subsequently been tried by M. Cutolo with interesting but somewhat ambiguous results.

(3) Measurement of separation in wave frequency of ordinary and extraordinary rays reflected from the same level in regions of high ion gradient. This experiment involves ready separation of the two rays at any wave frequency, using a

substantial refinement of the technique introduced by Ratcliffe and White.

All three methods require rather precise determination of height. Research on the third method is now under way at the Derwood Experimental Laboratory. Polarized receiving systems capable of quick frequency change are being developed as a first step in the elaboration of this technique. Designs are planned for precise measurement of frequency separation of echoes from the two wave components at any selected height.

Rocket methods. Closely related and complementary to the radio methods described above are direct measurements of the geomagnetic field at great heights by means of rockets.

In co-operation with the Applied Physics Laboratory of the Johns Hopkins University and the Naval Ordnance Laboratory, the earth's field was measured aboard the "Aerobee" rocket. A preliminary test flight at White Sands, New Mexico, on April 13, 1948 was completely successful. Continuous telemetered signals were obtained on both ascent and descent. The diminution in field with height was about 20 milligauss per 100 km, in good agreement with expectation from theory; a detailed check will be possible when more accurate information on the trajectory is available.

In subsequent experiments, it is hoped to determine the heights of electric current systems of the atmosphere responsible for the different geomagnetic variations.

Duplex-Channel Differential Recorder

The fundamental objective of this type of recorder is to provide the equivalent of a simple pen-and-ink record of the fluctuation of any dominant ionospheric characteristic, such as critical frequency, functions of virtual height, or absorption func-

tions of different layers. Present methods are extremely laborious, for they involve tedious scaling of thousands of multi-frequency records, and subsequent tabulation or graphical representation of the data for purposes of analysis. These multi-frequency methods do not automatically provide current information on events for the purpose of giving warning of unusual phenomena.

The basic idea underlying the new development involves essentially simultaneous transmission of radio waves on two channels separated by a small increment of wave frequency. The differences in characteristics of echoes on these two frequencies (in either amplitude or time delay, or both) provide the equipment with the information needed to contain the desired characteristic within the selected increment of wave frequency.

To utilize this information, certain independent decisions must be made by the equipment in comparing echoes on the two channels: (1) relative amplitude of echoes, (2) relative time delay of echoes, (3) selection of echoes of proper pulse shape and rejection of all other signals, (4) selection and integration of the appropriate number of successive echoes for the basis on which the comparison is to be made. If warning of unusual events is to be included, there must also be supplied (5) a source of information on normal events to provide the basis for comparison and warning, and a means of immediate comparison of the observation with the normal.

These decisions can be made by electronic circuits which in turn continually and automatically adjust the wave frequencies of the duplex channels to contain the selected characteristic. In like manner, other deciding elements of the circuit can be automatically adjusted. Thus the device "locks on and follows" the selected

ionospheric characteristic. Actual recording can be done from a voltage which is proportional to the shaft rotation (and, therefore, the wave frequency) of the basic oscillator, or to the parameters of other deciding elements of the circuit. Similarly, recording might be done on a magnetic tape permitting direct harmonic analysis and interpretation of the results without further treatment of the data.

Some of the critical elements of this device are now in the advanced-design stage, and the completed design will be tested during the ensuing year.

Experimental Cosmic-Ray Research

Large ionization chamber. As reported in the annual report of 1946-1947, a few sudden and unusually large increases in cosmic-ray intensity associated with great chromospheric eruptions were discovered by the Department as a result of continuous registration with Compton-Bennett meters of cosmic-ray intensity at several stations over a period of nearly ten years. These increases were probably due to magnetic acceleration of charged particles by changing magnetic fields of sunspots. This discovery provides the only evidence available for a mechanism by which a few of the cosmic rays may occasionally acquire their energies. It is, therefore, desirable to obtain as many observational data as possible on the phenomenon. Similar increases, though smaller or of shorter duration, may occur more frequently than those thus far observed. These increases may have escaped detection with the Compton-Bennett meters on account of the level of statistical fluctuations arising from the random arrival times of the primary particles. Since the level of these fluctuations in an ionization chamber can be decreased only by increasing the size of the chamber, a large ionization chamber of design similar

to that of the Compton-Bennett meters has been constructed for installation at Derwood. (See plate 2.)

The spherical Compton-Bennett chamber is about 1 foot in diameter. The new cylindrical chamber is 5 feet in diameter and 6 feet long. With this large chamber it is estimated that a change of about 3 per cent in cosmic-ray intensity should be detectable if the phenomenon lasts 5 minutes or longer. To be detectable in the Compton-Bennett meter a 3 per cent change would require about 4 hours. All instrument work for the large chamber has been completed, and the concrete supporting base has been constructed and lined with 4 inches of lead shielding. The chamber has been placed in position, and the housing for the instruments and recorders has been moved into place. The inside of the tank has been sand blasted to reduce alpha-particle contamination, and the tank filled with nitrogen (to prevent rust) under pressure to test for leakage. It is hoped to complete the assembly including the upper lead shielding and have the meter in operation by late summer. The 24 tons of lead shielding are on loan from the Office of Naval Research.

World-wide network of Compton-Bennett cosmic-ray meters. Operation of Compton-Bennett cosmic-ray meters was continued at Godhavn (Greenland), Christchurch (New Zealand), Cheltenham (Maryland, U. S. A.), and Huancayo (Peru). One meter which was being operated at Teoloyucan (Mexico) for Dr. A. H. Compton, at the time Professor of Physics, University of Chicago, was returned to the Department for a complete overhauling. This has now been effected except for reassembly and test at Derwood, where the meter will be used temporarily to determine the level of statistical fluctuations in the large ionization chamber.



Large ionization chamber for rapid measurement of fluctuations of cosmic-ray intensity (upper lead shielding not in place)

Another meter, which has been on temporary loan to the Physics Department at the University of Puerto Rico, is to be returned to the Department for overhauling. It is then planned to install it at Climax, Colorado, where, through the co-operation of Dr. Walter Roberts, Director of the Solar Observatory there, continuous

operation over many years is assured. The elevation of Climax is very nearly the same as that of Huancayo, making it an ideal location for determining the cause of the large systematic variation in burst frequency found at Huancayo and, to a lesser extent, at Teoloyucan, where the altitude is much less.

THEORETICAL AND STATISTICAL STUDIES

GENERAL REMARKS

The formulation of new problems and the continued interpretation of results of experiments received increased attention during the report year. A good beginning was also made on the theoretical work, which had been somewhat retarded because of the necessity of bringing to a conclusion considerable routine work relating to the older survey and observatory programs, and because major emphasis had to be placed on the initiation of a program of experiments. Progress was made chiefly along lines related to laboratory and field work. The advantageous interaction of experiment and theory was most apparent in the studies of cosmic rays and of the earth's crust. In the case of cosmic rays, welcome stimulus was afforded by the theoretical work of Vallarta, of Mexico, and Gill, of India, who joined with the staff of the Department as visiting investigators to collaborate in the analysis of the significance of the cosmic-ray increases associated with solar flares which were discovered here by Forbush two years ago. Plans were also formulated for work on specific problems of the upper atmosphere and solar phenomena, to be carried out with the help of Ferraro, of England, as visiting investigator during the coming year.

COSMIC-RAY RESEARCH

In last year's report the discovery of three unusually sudden and large increases in

cosmic-ray intensity was described. It was then suggested that these increases probably originated from charged particles which acquired their energies from a magnetic-accelerator action arising from changing magnetic fields of sunspots.

Subsequent investigations indicate that the rates of change of magnetic fields in large sunspot pairs or groups are sometimes capable of accelerating charged particles with the energies necessary for penetrating the earth's magnetic field at geomagnetic latitudes greater than 20° , where the cosmic-ray increases were observed. The required energies, of the order of 5 billion electron volts, appear to be available from the changing magnetic fields of some sunspot groups with associated intense flares, as indicated by Swann 15 years ago. Even so, a charged particle with energy of about 5 billion electron volts would normally be incapable of escaping from the sun, at latitudes where sunspots occur, on account of the blocking action of the sun's general magnetic field.

Sunspots, however, are regions of intense magnetic fields, and a pair of spots of opposite polarity comprises a transient magnetic dipole, designated M_s , for convenience, on the sun's surface. The sun's general magnetic field is equivalent to a dipole, M_e , at the sun's center and roughly antiparallel to the sun's axis of rotation.

As a consequence of M_e , there exist the well known forbidden regions of Störmer, across the boundaries of which no charged

particles with less than a specified energy can pass. The presence, however, of a sufficiently large sunspot dipole, in some orientations, creates a tunnel through which charged particles may escape across

open, it is necessary that the ratio of the magnetic field of M_{ss} to that of M_s , as a function of distance from M_{ss} , be changing sufficiently. Otherwise, there is effectively no tunnel all the way through the

inner forbidden region to the allowed region in which the earth lies (at a distance from the center of figure 4*A* about 6 times the length of the tunnel). Calculations of trajectories in the tunnel are still required to determine whether the above condition is also sufficient.

If these conditions prove sufficient, then an effective tunnel would have existed at the time of the largest increase in cosmic-ray intensity on July 25, 1946 (see figure). On the other hand, at the time of an intense solar flare on February 6, 1946, in the largest sunspot group yet recorded, calculations show that there was effectively no tunnel, and no cosmic-ray increase was observed (see figure). Effective tunnels probably existed also during the period of the cosmic-ray increases of February 28 and March 7, 1942, although this cannot be definitely stated until a more accurate determination of the direction of the M_{ss} involved can be made. The proposed mechanism thus explains why

a boundary of the otherwise forbidden regions to the allowed region, which, in figure 4, is the region between the two non-dashed curves shaped roughly like a circle and a parabola.

For the full length of the tunnel to be

cosmic-ray increases do not accompany all solar flares. To determine whether particles from the sunspots (if they escape through the tunnels) can reach the earth, trajectories from the sun to the earth still require calculation. The proposed mecha-

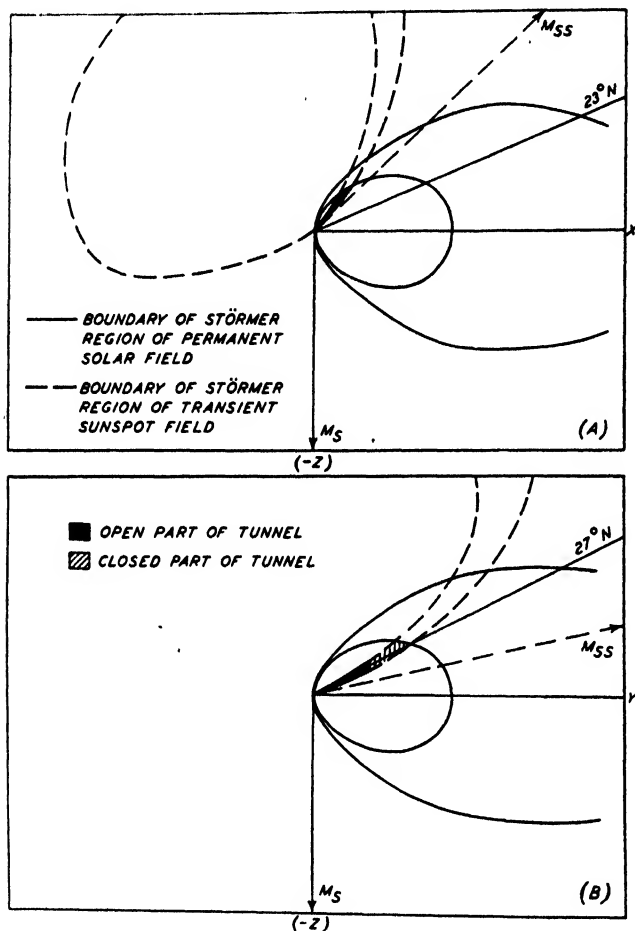


FIG. 4. *A*, tunnel on July 25, 1946, when large increase in cosmic-ray intensity was observed. *B*, no effective tunnel on February 6, 1946, when no increase in cosmic-ray intensity was observed.

nism appears quite promising for explaining how particles of cosmic-ray energy can be occasionally ejected from the sun. Incidentally, this is indirect new evidence for the existence of the general magnetic field of the sun.

The volume of bihourly values of cosmic-ray intensity at Huancayo (Peru) from June 1936 through December 1946, and the summaries of data through December 1946 for Cheltenham (Maryland), Christchurch (New Zealand), and Godhavn (Greenland), are in the hands of the printer. Several analyses based on these data have been completed and are nearly ready for publication. They include a study of the relation between magnetic disturbance and cosmic-ray intensity, reported at the meeting of the American Geophysical Union in the spring of 1948. This showed that the difference ΔC , representing the cosmic-ray ionization at Cheltenham, Huancayo, and Godhavn for the five international magnetically disturbed days minus that for the five international magnetically quiet days, was generally negative for each month, indicating some magnetic cut-off action during disturbed days. The average of ΔC over each year for the period 1937-1946 was always negative. Also, ΔC evidently follows the sunspot cycle, its absolute value being least during sunspot minimum. Similarly, the variation of daily means from the monthly means of cosmic-ray intensity at Huancayo was found to increase with sunspot activity.

Harmonic analyses of the 27-day and 13.5-day waves in cosmic-ray intensity for Cheltenham and for Huancayo were completed for all available data. The results provide material for statistical tests of the reality of such waves as would be expected at Cheltenham if the sun's permanent dipole were sufficiently inclined to the solar axis of rotation.

In order to determine the cause of an apparently anomalous increase, from 1937 to 1946, of about 6 per cent in the ionization at Huancayo, the data on bursts were critically studied. No significant variations were found in the frequency distribution of bursts according to size. Surprisingly, however, the rate of occurrence of bursts was found to have increased more than twofold between 1937 and 1946, paralleling the trend in ionization. No such change is found at other stations. Cosmic-ray records for Teoloyucan, Mexico, recently obtained from Dr. Compton, are now being analyzed, and preliminary results indicate a similar trend of smaller magnitude in the rate of occurrence of bursts at that station, the altitude of which is about 3000 feet less than that of Huancayo.

STUDIES OF THE EARTH'S INTERIOR

Spirited staff discussions, held twice weekly, with the participation of visiting colleagues, yielded effective group studies of problems of the earth's interior. Following formal lectures by Tatel, a number of problems were tentatively formulated in the field of isostasy, gravity, and seismology of the earth's crust. The most immediate benefits were felt in the clarifying of plans and objectives of the experimental program already under way for ascertaining by seismic means the constitution of the earth's crust, in both coastal and mountain areas. Studies of the earth's main field and its secular change were also made. In the coming year these discussions will be extended to include the upper atmosphere.

A theoretical study of the propagation of explosion waves within the earth's crust was begun. This work seems likely to be of considerable value in interpreting the experimental results of explosion tests and

wave observations in the seismic program, and in planning the locations of seismic receiving stations.

GEOMAGNETIC STUDIES

Plans were prepared in co-operation with the Applied Physics Laboratory and the Naval Ordnance Laboratory for measuring, by means of rockets, the heights of current systems flowing near the auroral zone during strong temporary intensifications of the geomagnetic field, known as bays. This could be done by noting magnetic changes observed as the rocket crosses the current layers. The height of the current system responsible for the solar daily variation will be sought near the equator. Calculations were made of the earth's main field to the top of expected trajectories, and of expected magnitudes of discontinuities in field on cross-

ing various current layers at different latitudes.

In preparation for further study of the association of solar flares with geophysical phenomena, such as cosmic-ray effects, ionospheric fade-outs, and magnetic storms, there was compiled a list of about 2000 flares during the period 1936 to 1947. It was found that of 160 most intense flares, 63 were followed by a magnetic storm within 4 days; and of 1500 moderately intense flares, only 53 were followed by a magnetic storm within 4 days. Hence, observable solar flares, although sometimes precursors of magnetic storms, are usually not followed by a magnetic storm within a few days.

In co-operation with the U. S. Coast and Geodetic Survey, a volume was published listing world *K*-indices of geomagnetic disturbance for the period 1940 to 1946, inclusive.

LABORATORY PHYSICS

NUCLEAR PHYSICS

The large pressure electrostatic generator has proved to be a very useful and reliable machine during the past year. After a number of improvements and repairs, the machine was in constant use for the last five months of the report year without breakdown. It operates reliably to 3.5 million volts, and observations have been made to 3.75 million volts. Voltage measurements are made to within ± 1000 volts, and the beam is homogeneous to within roughly ± 500 volts.

Major emphasis has been placed on the problem of the angular distribution and yield as a function of energy of the bombarding particle, for the alpha particles emitted in two reactions: lithium of mass 7 bombarded by protons, and lithium of mass 6 bombarded by deuterons. In each case the same compound nucleus, beryl-

lium of mass 8, is formed, which is unstable and breaks up into 2 alpha particles. The yield curves exhibit several resonances which are due to energy levels in the beryllium 8 nucleus. The angular distributions for both reactions can be represented by the expression $1 + A(E)\cos^2\theta + B(E)\cos^4\theta$. A study of the coefficients *A* and *B* revealed that *A* has a maximum in the reaction for lithium 7 at 1 million volts, and in that for lithium 6 at 2 million volts. The value of *A* for lithium 7 was observed to become negative above 2.75 million volts. *B* is negative for the lithium 7 reaction and positive for the lithium 6 reaction. Dr. Inglis has made a theoretical investigation of these reactions and has been able to fit the experimental angular distribution data with reasonable assumptions regarding the nuclei involved.

The energy-level systems for several

nuclei have been studied using reactions in which the deuteron is the bombarding particle and protons are emitted in the disintegration. The protons occur in a number of discrete energy groups the values of which give the energy levels in the residual nucleus. Sodium, aluminum, manganese, and carbon have been studied. Sodium and aluminum show respectively 12 and 15 proton groups with energies greater than that of the elastically scattered deuterons. The study of the oxygen proton groups last year and the carbon group recently has enabled us to distinguish these groups when observing with other elements, as both carbon and oxygen are usual contaminations for most targets.

Preparations have been essentially completed on a scattering apparatus to be used in observations on proton-proton scattering in the low-energy region between 200 and 500 kilovolts. Dr. Breit has pointed out that the scattering results in this energy region will be sensitive to the possible existence of a nuclear force between protons of longer range than that due to the abrupt and steep-walled potential well of great depth which is assumed in the analysis of proton-proton scattering at higher energies.

BIOPHYSICS

An area for research that gives real promise of new fruitfulness is a combined physical, chemical, and biological approach to the study of living matter. Too often in the past, compartmentation in science has interfered with progress in this important field. As a postwar development of the Department's long-continued and fruitful program in nuclear physics, and as a direct venture into fresh and challenging areas of research for physicists, the framework of a small but specially selected group has been assembled for

research in the problems of living matter. It consists of two biologists—one interested primarily in cell physiology, the second in biochemistry of enzymes, vitamins, and hormones—an organic chemist with special experience in biochemistry, and three physicists who, in addition to their own specialized work in nuclear physics and radioactive tracer work, have acquired experience in biochemistry and biology. Within the group is a wide and rich variety of technique, knowledge, and research experience, and each member is personally alive to the challenge set by the remarkable organization shown in the physicochemical behavior of living systems. Since the group is small, there is a large degree of cross-fertilization and self-training. Thus, for instance, the physicists are constantly learning from the biologists new aspects of the behavior of living matter, while the physicists help the biologists with the key facts and possibilities of nuclear physics and the physicochemical approach.

The result of this blending has been that all the members of the group have been greatly stimulated in their researches. As part of the training program, one physicist spent two months at the Marine Biological Laboratory at Woods Hole, and a second physicist spent a similar period at the Department of Genetics of Carnegie Institution at Cold Spring Harbor. The strength of the group is further reinforced by contact with numerous collaborators in the Washington area. Through individual efforts, team work within the group, and collaboration with outside research workers, numerous studies have been carried out during the past year. Among the results of projects under way or reaching completion are the following:

Studies of capillary permeability and placental permeability to electrolytes and proteins (in collaboration with Louis B.

Flexner, of the Institution's Department of Embryology, and Gilbert J. Vosburgh, of the Department of Obstetrics, Johns Hopkins University and Hospital) have thrown new light on the mechanisms of the capillary bed and on means by which the embryo obtains needed metabolites.

Study of the fate of stibine taken in by the respiration has demonstrated an interesting method by which this compound is detoxified by oxidation steps catalyzed by hemoglobin. Robert E. Smith, J. Murray Steele, and Robert E. Eakin, of the Naval Medical Research Institute, collaborated in this study.

Investigation (with Roy O. Scholz, of the Wilmer Institute, Johns Hopkins University and Hospital) of the turnover of Na^{22} in aqueous humor has demonstrated the very rapid equilibrium of this substance with Na^{22} in plasma.

Further studies in collaboration with William R. Duryee, of the National Cancer Institute (stationed at our Department), of permeability and diffusion in the frog's egg have demonstrated extremely rapid "diffusion" of K^{42} once that substance is inside the egg. This work yields a crude estimate of the diffusion coefficient of potassium within living matter, which is at least as high as that in an ordinary aqueous solution.

Studies have also been made by Duryee on some effects of radiation on living matter. It has been shown that both beta-radiation (P^{32}) and X-rays produce a striking phase change in the nuclei of eggs of frogs and salamanders. Another study indicates that one effect of radiation on *Escherichia coli* is a disturbance of a particular enzyme balance.

Another series of experiments has been directed to a study of the P^{32} metabolism of bacteria (*E. coli*) as a function of the state (growing or resting) of the organism. The delay in virus growth after ultra-

violet radiation observed by Luria was confirmed, and additional information on the process was obtained.

Development (here and at Johns Hopkins Hospital by Duryee) of a microdissection technique has made available for various studies good quantities of unfertilized human ova.

As a more detailed illustration of the type of studies carried on by the group, the two following investigations are cited.

Since phosphorus is known to be crucial in the energy utilization of living matter, a study of phosphorus take-up in dividing cells is of some importance. The eggs of the sea urchin *Arbacia punctulata* provide suitable biological material for such a study. They can be obtained in large quantities and fertilized at will. The result is a relatively uniform group of cells, almost all of which divide on a definite schedule.

Studies have been made of the permeability of the unfertilized and fertilized eggs. In the case of fertilized eggs, experiments were made both at 10° and at 23°C . In addition, the effect of a metabolic poison, 4-6 dinitro-o-cresol, was observed. The results showed that phosphorus take-up of the eggs is quite closely connected with cellular activity and can serve as a more precise measure than has hitherto been available. Thus, the amount of phosphorus taken up by the fertilized eggs was 40-fold greater than that taken up by unfertilized eggs. It is interesting to note that oxygen metabolism has been found to differ by only a factor of 3 in this case, and even resting cells consume oxygen at a fair rate. At 10°C the amount of phosphorus found in the eggs was only one-seventh that found for a similar exposure at 23°C . As is well known, the relative times for first cleavage differ by the same factor. When 1.6×10^{-5} molar 4-6 dinitro-o-cresol is present, oxygen me-

tabolism is about doubled and cell division is almost stopped. Under these circumstances phosphorus take-up was diminished by a factor of 6. A survey of the fate of the radioactive phosphorus taken in at 23° C showed that most went into the "acid-soluble fraction" (phosphorus compounds soluble in 5 per cent tri-chlor acetic acid). With short exposures to radioactive phosphorus, the "acid-soluble fraction" (largely related to cellular energy production) contained 10 times as much activity as the "acid-insoluble fraction" (largely related to protein synthesis). Most of the "acid-soluble fraction" was in the form of organic phosphate. One interesting feature was that the phosphorus uptake did not increase immediately after fertilization. A transition period of 7 to 10 minutes elapsed during which a new level of phosphorus intake was attained.

A second example of biophysics work in progress at the Department is a study of calcium metabolism by Hugh H. Darby, Visiting Investigator. The production of calcified structures in the animal kingdom has been sporadic. There have been calcium shells in the protozoa, calcium deposits by the corals and the sponges, the calcium of the crustacean exoskeleton, and the calcium shells of the mollusks, but it was only when evolution reached the vertebrates that a really definite internal calcium skeletal structure was developed. This skeletal structure has made possible a size and diversity in the vertebrates that is distinctly dependent on it.

The researches of the late '20's and early '30's resulted in the recognition of the role of one substance in the forming of bone in the developing animal; namely, vitamin D. The Carnegie Institution took part in the work done on the origin of "D" at the Tortugas Islands in the middle '30's. The work on vitamin D, however, led to no fundamental answer on how calcium was

laid down in the bones, and attempts to discover the mode of operation of vitamin D in the laying down of calcium gave negative results. The topic was dropped as a subject of research.

There remained the following problems, however. First, there was no knowledge of how, when, or why the calcium reached the bones. There was no indication, with or without vitamin D, of the increase in blood calcium during the time when bone was forming. The problem of repair of bones in adults was also untouched, because experimental feeding of large quantities of vitamin D led to no enhancement of healing in the broken adult structures. Researches were started in our laboratory, therefore, to find out (1) how calcium reaches the bones, and (2) whether it is possible to find somewhere in the body a controlling influence on bone metabolism. The previous findings on the relation of the thyroid and parathyroid to calcium were well known, but had never been applied to bone formation. The fact that the thyroid closely antedates in phylogenetic history the formation of bone in the fishes seemed to warrant careful study.

It has now been shown by using radioactive calcium in normal animals that the calcium enters the bone even at the sites of least calcification, but that its stay there is very temporary and in 72 hours some of the calcium has already left these sites. This means that there must be some other system causing the calcium actually to deposit and form bone. It also shows that the metabolism of calcium is equal to the metabolism of other materials, as found by Schoenheimer and Rittenberg many years ago. A rapid turnover is indicated in all metabolites, even the heavy metals.

On the removal of the thyroids and parathyroids in the rat, the calcium reaches the bones just as before in a very short time, but it leaves the bones almost im-

mediately. This gives a clue to the role of the thyroid in the deposition of calcium in bone.

Destruction of the thyroid by chemical methods is possible; for example, thiouracil causes enlargement and destruction of the thyroid. It had been noticed also that vitamin D deficient rats had enlarged thyroids, but no research had been undertaken to ascertain the cause. By destroying the thyroid with thiouracil and counteracting this with vitamin D, the relation of that vitamin to thyroid metabolism was definitely established. The role of the thyroid in causing calcium deposition in the bone is now open to further research. Following this discovery of the relation of vitamin D and thiouracil, investigation of rats with only thiouracil medication showed interesting changes

in the laying down of calcium at the epiphysis. Since the origin of the teeth is phylogenetically similar to that of bone, some preliminary experiments with similar results have been made on the teeth of rats. An opening wedge has been driven into the problem of the formation of bone and teeth which seems to have wide application.

During the report year very good progress was made in increasing the laboratory space and equipment. Four additional laboratory rooms were made available for biophysics work. Much useful equipment has been produced, including five complete Geiger counter sets. A mass spectrograph is nearing completion. It will permit the use of stable isotope tracers to supplement the radioactive tracer work for special problems.

OPERATIONS AND STAFF

CO-OPERATIVE WORK OF THE DEPARTMENT

The Department has continued the policy of advancing projects through co-operation with individuals at home and abroad and with governmental and other organizations.

Research and development work on the deeper layers of the earth's crust was continued under Contract N70nr-290 with the Office of Naval Research. Another contract, N70nr-459, dated November 1, 1947, with the same office provides for Navy co-operation in our cosmic-ray studies. A third contract, NOa(s)9318, with the Navy was negotiated on November 18, 1947, and provides for co-operative research and development directed toward the improvement of airborne magnetometers. All these contracts are written on a no-fee or \$1.00-fee basis.

Assistance was given, principally by the loan of instrumental equipment and granting of necessary leaves of absence,

to two members of the Department's staff in order that they might undertake a classified contract with the War Department. Various members of the staff have also assisted governmental establishments as consultants; one has been on a full-time leave-of-absence basis from February 1, 1948.

Our former observatories in Australia and Peru were furnished special equipment and supplies not readily obtainable locally. Books and scarce electronic supplies were sent to Dr. Kwei in China and Professor Amaldi in Italy.

Through a special appropriation by the Institution, arrangements were made to supply Dr. Hume-Rothery in England with vanadium metal of high purity for metallurgical research.

The Department has had the advantage of collaboration with Dr. Louis Flexner, of the Department of Embryology, in the biophysics program, and with representa-

tives of the National Institutes of Health, the National Bureau of Standards, the Applied Physics Laboratory of the Johns Hopkins University, the Brookhaven National Laboratory, the Wilmer Institute and the Department of Obstetrics of the Johns Hopkins University, the University of Texas, and Tulane and Howard Universities.

In addition, the biophysics group has been responsible for the operation of the cyclotron. This has continued to be useful in the production of radioactive isotopes, which have been distributed free of charge to some forty different groups both in this country and in Australia, Canada, England, New Zealand, South Africa, and Sweden. Special bombardments were carried out for the Clinton and Brookhaven laboratories.

In the investigation of sudden increases in cosmic radiation associated with solar flares, the Department has had the advantage of collaboration with Dr. M. S. Vallarta, of the Comisión Impulsora y Coordinadora de la Investigación Científica, e Instituto de Física, Universidad de México, and of P. S. Gill, of Tata Institute of Fundamental Research, Bombay, India.

In the seismic program, close collaboration and joint experiments were carried out with Columbia University and the New Mexico School of Mines, as well as with Navy and Army groups. The seismic program is entirely open and unclassified, as is every other item of the Department's work. Grateful acknowledgment is made to the Navy (Office of Naval Research and Bureau of Ordnance) for numerous underwater explosions scheduled for the convenience of our observers, and to various quarry operators in Virginia and Pennsylvania who made arrangements for us to time accurately their blasting operations.

THEORETICAL PHYSICS CONFERENCE

This series was continued with a Tenth Washington Conference on Theoretical Physics in Washington, D. C., November 13-15, 1947, under the joint sponsorship of the George Washington University and the Carnegie Institution of Washington. The subject selected was "Gravitation and electromagnetism." The discussions by some twenty-four representatives of universities, research organizations, and government bureaus covered general problems of the expanding universe, the Schroedinger and Blackett proposals regarding the relation between gravitation and electromagnetism, and topics relating to the unified field theory. One special topic of discussion, in response to questions under study by the Department, concerned the possibility of an experimental search for the degradation of frequency of light waves by some physical mechanism which might operate during passage through long reaches of space (the "red shift"). The questions remain interesting and perhaps not without avenues of approach.

PUBLICATIONS

Aside from the scientific papers appearing in the regular literature during the year, there have been published seven volumes of the Researches of the Department, containing the results of observations of terrestrial magnetism and of the ionosphere. There was also issued a special volume dealing with the description and analysis of the geomagnetic field. All these volumes are listed under "Major publications" in the bibliography at the end of this report. Moreover, there will be published in 1948 a volume dealing with cosmic-ray results from Huancayo Observatory, Peru, June 1936-December 1946, and including summaries through

1946 from observatories at Cheltenham, Christchurch, and Godhavn.

The following publications, prepared for offset printing, have been, or will be, microfilmed, and the negatives will be kept on file at the Department in order that positives may be supplied to an international list of interested agencies and to others on request. It is proposed to publish these volumes at some future date, depending on the demand for data and other factors. Pending publication they are to be regarded as Department publications in microfilm. CIW Publication 175, volume IX, "Earth-current results at Tucson Magnetic Observatory 1932-1942," has already been microfilmed, and the following are to be microfilmed during 1948: volume XV, "Earth-current results from Huancayo Observatory 1932-1942"; volume XVI, "Earth-current results from Watheroo Observatory 1932-1942"; volume XVII, "Atmospheric-electric results from Watheroo Observatory 1924-1934"; volume XVIII, "Atmospheric-electric results from Tucson Magnetic Observatory 1931-1934"; and volume XIX, "Atmospheric-electric results from Huancayo Observatory 1924-1934."

ADMINISTRATION AND OPERATION

A new heating plant was installed in the main building, and the former coal vault was converted into a measurements room for the use of the biophysics group. Fire walls and doors were added in the attic for the protection of archives. The main shop was rearranged and steel shelving

added, and similar installations were made at the Derwood Experimental Laboratory. Plans were made and initial steps taken to beautify the grounds.

To provide for ever-increasing demands for transportation concerned with research activities, a new Ford sedan and surplus radar truck were purchased. A panel truck was borrowed from the Navy for use in our seismic work.

Final report and vouchers were submitted to conclude the one remaining war contract which carried over into this report year.

The *Journal of Terrestrial Magnetism and Atmospheric Electricity*, edited by Dr. John A. Fleming, retired Director of the Department, was given continued support.

The Department again had the privilege of visits from a number of guest investigators for varying periods. These are shown in the appended list, along with the regular members of the staff, most of whom were employed for the entire report year.

LECTURES GIVEN BY NONMEMBERS OF THE DEPARTMENT

November 21, 1947, "The mechanics of mountain building," by Ross Gunn.

March 2, 1948, "First products of photosynthesis," by William Fager.

March 12, 1948, "Theory of ion exchange," by George A. Boyd.

May 10, 1948, "The origin of the earth's main field." This was a discussion by staff members in which G. Gamow, Ross Gunn, and other visitors participated.

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STAFF AND ORGANIZATION

SCIENTIFIC STAFF

Staff Members (new title, from January 1, 1948):

Geophysics: L. V. Berkner, S. E. Forbush, O. H. Gish, R. W. Goranson (from Geophysical Laboratory), E. A. Johnson (on leave of absence for governmental research), W. J. Rooney, H. E. Tatel, O. W. Torreson, M. A. Tuve, E. H. Vestine, G. R. Wait, H. W. Wells.

Laboratory and Biophysics: P. H. Abelson, D. B. Cowie, N. P. Heydenburg, R. B. Roberts, M. A. Tuve.

Guests, Associates, Fellows, and Visiting Investigators: S. Banerjee, C. S. I. R. Laboratories, Old Delhi, India; G. E. Boyd, Oak Ridge National Laboratory; M. Casaverde, Geophysical Institute of Huancayo, Huancayo, Peru; C. Y. Chao, National Central University, Nanking, China; H. H. Darby; Miss E. Dollman, Brookhaven National Laboratory; W. R. Duryee, National Institutes of Health; P. S. Gill, Tata Institute of Fundamental Research, Bombay, India; J. W. Graham, Johns Hopkins University; E. J. Habib, Catholic University;

E. M. Hafner, Brookhaven National Laboratory; R. Hossfeld, Catholic University; C. M. Hudson, Office of Chief of Ordnance, War Department; D. R. Inglis, Johns Hopkins University; E. E. Motta, Oak Ridge National Laboratory; T. Murphy, University College, Dublin, Eire; A. T. Ness, U. S. Public Health Service; W. D. Parkinson; T. H. Pi, National Central University, Nanking, China; Miss J. Roquet, Institut de Physique du Globe, University of Paris; J. E. Sreb, Applied Physics Laboratory, Johns Hopkins University; F. L. Talbott, Catholic University; M. S. Vallarta, Instituto de Física, Universidad de México, México, D. F., and Comisión Impulsora y Coordinadora de la Investigación Científica, México, D. F.; W. D. Whitehead, Jr., University of Virginia.

OPERATING STAFF

Administrative: M. B. Smith, W. F. Steiner.

Office and Clerical: Mrs. J. H. Campbell, J. J. Capello, Miss R. C. Dermody,* W. N. Dove,

H. D. Harradon, W. C. Hendrix, Miss H. E. Russell, A. D. Singer.

Instrument Shop: B. J. Haase, L. A. Horton, J. G. Lorz, F. B. Thomas.*

Research Assistants, Laboratory Assistants, and Technicians: S. J. Buynitzky, J. B. Doak, D. H. Gleason,* P. A. Johnson, C. J. Ksanda, P. G. Ledig,* C. A. Little, Jr., P. F. Michelsen, P. L. Moats, A. E. Moebs, W. C. Parkinson, W. D. Parkinson,* R. W. Reuschlein, W. E. Scott.

Computers: Miss E. Balsam, Miss I. Lange.

Maintenance: C. Balsam, C. W. Burger, C. Dom-ton, S. W. Malvin, E. Quade.

Part-Time and Temporary Employees: There were 23 part-time and temporary employees and students engaged during the year, usually for short periods, to assist in the office and laboratory work. Miss M. Walburn, production assistant, was assigned by the Office of Publications to publication work at the Department to May 25, 1948.

* Resigned.

SPECIAL PROJECTS: TERRESTRIAL SCIENCES

JOSEPH C. BOYCE, College of Engineering, New York University, New York, N. Y.
Research in the spectroscopy of the vacuum ultraviolet. (For previous reports see Year Books Nos. 38 to 41.)

Following the war, the Carnegie vacuum spectrograph was moved from the Massachusetts Institute of Technology to New York University, together with the library of plates and other records of the project. The spectrograph itself has not yet been reassembled, since a considerable quantity of data obtained before the war has still not been reduced.

The first opportunity to resume investigation came in the spring of 1947. Since that time data on nickel, cobalt, and molybdenum have been reduced to wave-length and wave-number lists. Copies of these lists have been furnished to the National Bureau of Standards, where analysis of these elements is in progress. The nickel list is currently being searched here for regularities which might be ascribed to the third spectrum of that element.

Line spectra no longer present the challenge to investigation which they did even a decade ago. The principal features of most spectra have been recognized, and a representative number of spectra are known in considerable detail. Young physicists no longer are attracted by the remaining problems. Under these circumstances there is danger that some existing knowledge may be lost. A project has recently been initiated at the National Bureau of Standards for collecting and publishing all existing information on atomic energy levels as derived from anal-

ysis of spectra. These tables will bring together, with a consistent notation, not only previously published results, but many additional fragments which would not have justified separate publication.

Wherever vacuum ultraviolet data from the records of our own project can assist in completing these tables, such data will be forwarded to the National Bureau of Standards. The work done this year on nickel, cobalt, and molybdenum was undertaken for this purpose.

It is pleasant to record continuing co-operation with Professor B. Edlén, of Lund, Sweden. Plates have been lent to him covering the spectra of a number of lighter elements, up to calcium, in the wave-length region from λ_{2500} to λ_{1200} . These will supplement spectra taken in his laboratory at both shorter and longer wave lengths.

Vacuum ultraviolet spectra of three heavy elements have been lent to Professor C. J. Bakker, of the Zeeman Institute in Amsterdam, to be used in connection with measurements made at longer wave lengths in that laboratory.

A spectrogram of manganese has been lent to Dr. Catalan, currently at the National Bureau of Standards, to assist in his analysis of the spectrum of that element.

Mrs. Bess Vogel was employed as a research assistant from June 1947, her appointment to terminate in August 1948.

DIVISION OF PLANT BIOLOGY

Stanford, California

C. STACY FRENCH, *Director*

The fundamental question of how plants make food, central in the Division's investigations during the chairmanship of Dr. Herman A. Spoehr, continues to be the prime focus of research. Dr. Spoehr became chairman of the Division in 1928, and during the nineteen years of his administration the activities of the Division were consolidated, a modern laboratory was constructed, and an able staff was assembled and guided through a long period of intensive research. In this period, basic studies of the nature of the process and products of photosynthesis and the interrelationships of plants with each other and with their environment greatly elucidated these aspects of biology. The broader aspects of the fields which have constituted the work of the Division through these years continue to be of the greatest significance in relation to present scientific knowledge.

In study of the fundamental food-making process in plants, both the well established and also some newly opened approaches that should lead to a deeper understanding of the phenomenon are being used. Dr. Spoehr, now freed of administrative burdens, is vigorously continuing his researches on the products of photosynthesis, yet gives generously of his time in connection with the work of the rest of the staff.

During the year one group of investigators, dealing with the biochemistry of plants, has attacked various problems closely concerned with the process of photosynthesis, while the experimental taxonomy group has been occupied with further work on the relationships of plants, spe-

cifically on the study of ecological genetics of climatic races within a single species. The latter investigators have also been following up the purely scientific aspects of the range-grass breeding program that began as a war project with primarily practical interests.

The means by which the green plant is able to convert energy from sunlight into chemical energy in the form of food available for plant and animal growth is one of the more fundamental processes of life. There exists a large amount of specific information about photosynthesis, but, nevertheless, there has been no clear demonstration of the identity and sequence of all the separate steps that are involved. Our specific approaches toward the clarification of these steps are along separate but interrelated lines of endeavor.

Pigments participate in photosynthesis by their absorption of light. One of the first observable chemical effects of this absorbed light is the splitting of water molecules. Pure pigments alone do not split water molecules with the simultaneous evolution of oxygen in this way. This step of photosynthesis is, however, carried out by isolated chloroplasts even when they are disintegrated. It is, therefore, evident that for this step of photosynthesis, substances, probably enzymes, are required in addition to pigments. These other essential components of the photosynthetic apparatus are as yet of unknown composition and properties. By the study of various fractions from finely disintegrated chloroplasts, it may be possible to find out something about the nature and function of the substances which, in addition to the

pigments, are responsible for the activity of chloroplasts. Progress depends on the devising of suitable techniques for the separation of such components.

During the past year the procedures for the preparation of disintegrated chloroplast material and the measurement of its activity have been improved; equipment has been designed and constructed for this purpose. A large number of preparations of disintegrated chloroplasts have been fractionated and the activity of these fractions has been tested.

The development of chlorophyll in the early stages of plant growth is being investigated. Plants which differ by heredity in their ability to form chlorophyll provide valuable material for this purpose. The simultaneous elaboration of the pigments and other parts of the photosynthetic apparatus is being followed by observation of the activity of chloroplasts obtained from plants at various stages of development.

Another line of approach has been the investigation of the chloroplast pigments in regard to their distribution in different plants. A fundamental relation between the phylogenetic classification of various species of plants and the type of pigment complex found in them has been further elucidated.

A tremendous diversity of form and physiological characteristics may be contained within a single species. An experimental approach to the understanding of the origin of such great diversity has been followed in studies on the climatic races of *Potentilla glandulosa*, a member of the rose family. Four of the subspecies of this plant exhibit great differences in their range of tolerance of widely different environmental conditions and in their morphology. Each one is particularly well adapted to its native environment. Crosses

between them have been made and the progeny grown under various conditions, both in the gardens at Stanford and at the stations in the mountains. Those physiological characters which determine the plant's ability to thrive under given climatic conditions are usually found to be linked to various morphological characters that determine its appearance. The hybrid derivatives obtained from crossing the subspecies recombine both the physiological and the morphological characters in almost all possible ways. Such an experiment demonstrates conclusively that there are very many possible genetic combinations which have not yet appeared in nature owing to the geographic isolation of potential parents. Also it shows that the hybrids between parents from widely different climates may sometimes yield hardier and more vigorous progeny than any existing type, even within the range of the parents. A conclusion to be drawn from these experiments is that many possible lines of evolution are still untried in nature and that evolution is therefore a continuing process that has by no means reached its ultimate limits.

The relative merits of a number of the more promising range-grass hybrids of the genus *Poa* have been compared at the three transplant stations of the Division; some of these hybrids were turned over to the Soil Conservation Service of the U. S. Department of Agriculture, and are being tested in two of their nurseries. Members of the Division staff are following these trials. Some of these strains are now ready for more extensive testing under field conditions, and, if suitable, will eventually be made more widely available.

Another possible application of fundamental investigations to the potential food supply of the world is the development of means by which the content of fats, pro-

teins, and carbohydrates in a plant may be varied at will within wide limits by a choice of appropriate conditions for growth. *Chlorella*, long a favorite organism for the laboratory investigation of photosynthesis, can be grown under con-

trolled conditions to produce a high content of fat or protein. Despite technical obstacles there is reason to believe that the feasibility of doing this on a scale large enough for practical purposes should be tested.

PERSONNEL

ADMINISTRATION

C. Stacy French, *Director*
Wilbur A. Pestell, *Secretary*
James W. Groshong, Tieh-Ying Wu, *Custodians*

BIOCHEMICAL INVESTIGATIONS

Staff: Harold W. Milner, James H. C. Smith,
Herman A. Spoehr, Harold H. Strain
Fellows: Violet M. Koski, Fergus D. H.
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Research Assistants: Marie L. Koenig, Henry C.
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Staff: Jens C. Clausen, William M. Hiesey,
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Research Assistants: Helen K. Sharsmith, Robert W. Ayres
Gardener: Wesley B. Justice

RESEARCH ASSOCIATE

Ralph W. Chaney (Professor of Paleontology,
University of California, Berkeley, California)

BIOCHEMICAL INVESTIGATIONS

THE PHOTOCHEMICAL ACTIVITY OF ISOLATED CHLOROPLASTS

C. S. FRENCH, H. W. MILNER, M. L. KOENIG,
AND F. D. H. MACDOWALL

The conversion of light energy to chemical energy by photosynthesis involves the decomposition of water. Oxygen is given off as a gas, while the hydrogen from the water is used for the formation of reduced substances by illuminated chloroplasts. The utilization of these reduced substances in the reduction of carbon dioxide is under intensive investigation in other laboratories with the use of radioactive carbon.

We have been concerned primarily with the initial stages of photosynthesis, that is, with the reducing action of chloroplasts, which is brought about by the absorption of light by the chlorophyll contained in them. It is this reducing ability of chloroplasts which is fundamentally characteristic of green plants. Many organisms are able to reduce carbon dioxide provided they are artificially supplied with a re-

ducing substance. Only in green plants is this reducing ability produced directly by light.

Though the complete process of photosynthesis, that is, the reduction of carbon dioxide, does not take place in isolated chloroplasts removed from the living plant, they nevertheless show a reducing activity in themselves on being illuminated. Since this activity is not destroyed by removing chloroplasts from the living plant, or even by their thorough disintegration, it appears that this is one of the more promising means of studying the functions and properties of discrete parts of the photosynthetic system. This is true because the activity may be obtained in an extract, and is, therefore, more amenable to preparative biochemical investigation than are other parts of the photosynthetic apparatus. Our objective is to distinguish in disintegrated chloroplasts those components which are essential to the reduction process; to find out, if possible, what substances are thus

actively concerned in the fundamental conversion of light energy into chemical energy in the form of a reducing compound; and to clarify the relation of this process to carbon dioxide reduction. The evidence for the reducing activity of extracts from chloroplasts is found in their reducing action on substances other than carbon dioxide. For the purpose of measuring the reducing activity of our preparations, we have used the dye 2,6-dichlorophenol indophenol, which in the oxidized state is blue, but which under the action of illuminated chloroplasts is rapidly reduced to the colorless state. The activity may conveniently be measured by following the color change of the dye. Thus it is possible to express quantitatively the reducing activity of preparations made from chloroplasts. By this means we have studied the activity of fractions obtained from disintegrated chloroplasts.

Supersonic vibration has been the most satisfactory means which we have encountered for this disintegration. It breaks up chloroplasts into extremely small particles, possibly even into individual protein molecules. Upon extended centrifugation at high speed they still remain in suspension or solution, giving clear green preparations which retain the reducing activity of the chloroplasts from which they were obtained. This material, however, like preparations of isolated chloroplasts themselves, is unstable and must be stored and handled at temperatures just above freezing in order to reduce the rapid loss of activity.

More reliable methods for preparing disintegrated chloroplast material and for decreasing its rapid loss of activity are being sought. Chloroplasts are separated more easily from leaves which have been subjected to a temperature just low enough to freeze them. More severe freezing of the

leaves is detrimental. Freezing of the isolated chloroplasts results in a loss of some of their activity. It has been found that preparations of disintegrated chloroplasts are inactivated by having gases bubbled through them, by the presence of oxygen, or by light. The loss of activity is slowest at a temperature just above freezing, for which reason as much as possible of the work is done in a cold room at 0° C. Though potassium chloride has some stabilizing effect on the intact chloroplasts, its use leads to slow precipitation of the disintegrated chloroplast material and is, therefore, to be avoided if possible. Ethylene glycol, propylene glycol, and glycerine stabilize somewhat, but their presence complicates the nitrogen analyses.

During the loss of activity in storage at 0° the chlorophyll changes, as has been shown by the chromatographic separation of methanol extracts. In addition to pigment changes, it is likely that there are also other factors concerned with the instability of chloroplast preparations. In some cases chloroplast material stored at 0° C. in the dark has, in the absence of oxygen, retained up to one-third of its photochemical activity for as long as 2 weeks. The most rapid loss of activity occurs in the first 24 hours. The addition of common antioxidants has only a very small effect in stabilizing the activity. Potassium cyanide neither affects the activity of the fresh material nor acts as a stabilizer. The instability of chloroplast preparations still causes the greatest technical difficulty connected with this work.

Standard chemical methods for nitrogen and chlorophyll determinations have been modified in order to measure these components rapidly and accurately in small amounts of material. The activity measurements may also be made with extremely small volumes of the preparation.

Fractional precipitation of the disintegrated chloroplast material by salts, such as ammonium sulfate, or sodium sulfate, gives preparations with some activity. These preparations always have less activity than the starting material and show no clear-cut separation of fractions containing more active material. The best method of precipitation that we have found is the use of very dilute acid buffers. By this means precipitates have been obtained which, when resuspended, give activities about equal to that of the starting material. Even with this procedure we have not been able to concentrate fractions having appreciably more activity than the initial extract. Methods of partial precipitation of the chloroplast material are being investigated further. Fractional precipitation by the addition of alcohol, which has yielded such excellent results with blood serum, has not been applicable as yet to disintegrated chloroplasts, since they contain enough fat to protect the protein components from precipitating even in very high alcohol concentrations.

Chlorophyll is perhaps the most obvious organic substance in the world, since the predominant color of natural vegetation is due to its presence. Its chemistry in the natural state is, however, only crudely understood. Chlorophyll which has been extracted by organic solvents certainly is a different substance from natural chlorophyll in leaves. This is indicated by the shift in its absorption spectrum and the changes in its other properties. The chemistry of this extracted chlorophyll has been well worked out. The condition of chlorophyll in leaves has long been thought to be somewhat similar to that of hemin in the blood, in which it is combined with protein to form hemoglobin. The chemical properties of hemoglobin have been studied intensively, and its chemical nature is well

known. Natural chlorophyll has been given the name chloroplastin; its properties have, however, been extremely difficult to investigate, since the protein to which the chlorophyll is presumably attached is generally insoluble in water, and therefore preparations of pure chloroplastin are hard to obtain. This material contains a large amount of fat, which greatly complicates the chemical separation of the still largely hypothetical chlorophyll-protein compound.

The reducing activity of isolated chloroplasts depends upon light absorbed by chloroplastin and probably upon enzyme action. The preparation in a pure form and the determination of the chemical composition of chloroplastin is obviously one of the several problems concerned in the study of the reducing activity of isolated chloroplasts. By making analyses for the nitrogen, chlorophyll, and fat content of various fractions obtained from disintegrated chloroplasts, it may be possible to find fractions which contain this substance predominantly, for the further investigation of its chemical composition and its relation to the other components of the photosynthetic apparatus of living cells.

CHLOROPHYLL FORMATION

JAMES H. C. SMITH AND VIOLET M. KOSKI

An analysis has been made of the processes by which chlorophyll is formed in plants by examining the effects of light and temperature on seedlings which vary in their inherent ability to become green. Barley seedlings, which are yellow when grown in the dark, become green when placed in the light. Pine seedlings are green even though they are germinated in complete darkness. Albino corn seedlings are nearly colorless when grown either in the dark or in the light.

It has been reported in the two previous

Year Books (No. 45 [1945-1946], pp. 104-106; No. 46 [1946-1947], pp. 88-90) that chlorophyll appears to be derived from preformed organic compounds containing magnesium—especially protochlorophyll. During the past year quantitative determinations have been made of the changes taking place in the protochlorophyll and chlorophyll content of dark-grown barley seedlings during subsequent illumination at low temperatures. Under these condi-

solid finally obtained, 2.73 per cent. A sample of the purest solid protochlorophyll obtained thus far, when dissolved in ether, acetone, or methanol gave the values shown in table 1 for the specific absorption coefficients and the wave lengths at the absorption maxima. Included also in table 1 are values for an ether solution of protochlorophyll in which the pigment had not been precipitated and dried. The concentration of the solution was calculated from

TABLE 1

THE SPECIFIC ABSORPTION COEFFICIENTS AND THE WAVE LENGTHS FOR THE ABSORPTION MAXIMA OF PROTOCHLOROPHYLL

PRECIPITATED AND DRIED						UNPRECIPITATED	
ETHER		ACETONE		METHANOL		ETHER	
λ max. ($m\mu$)	α^*	λ max. ($m\mu$)	α	λ max. ($m\mu$)	α	λ max. ($m\mu$)	α
623.....	36.9	623.....	34.9	629.....	27.2	623.....	39.9
571.....	14.0	571.....	13.5	578.....	10.5	571.....	14.9
535.....	6.8	535.....	6.9	535.....	7.2
432.....	305.9	432.....	270.5	434.....	177.3	432.....	325.5

* $\alpha = \frac{1}{lc} \log_{10} 1/T$, where l = cm., c = gm./liter, and T = fraction of incident light transmitted.

tions there is a one-to-one correspondence between the amount of chlorophyll which is produced and the amount of protochlorophyll which disappears.

Measurements of this transformation were made possible by a determination in absolute units of the spectral absorption of protochlorophyll. The protochlorophyll used for the spectroscopic determinations was extracted from barley leaves which had been grown in continuous darkness. After extraction, the protochlorophyll was isolated and purified by a series of adsorptions and precipitations. The high degree of purity of the product was attested by the magnesium content of the amorphous

magnesium analysis. Comparison of the absorption values shows that precipitation and drying lowers the coefficients to some extent.

Our measurements have shown the absorption curve of protochlorophyll to resemble the over-all action spectrum curve for chlorophyll formation. The correspondence of these two curves strongly indicates that protochlorophyll is the light-absorbing agent concerned in the initial formation of chlorophyll, in addition to being the actual substance from which chlorophyll is formed.

The quantity of chlorophyll produced by the transformation of the protochloro-

phyll initially present in leaves is only a small fraction of the total amount which appears upon extended illumination. For protochlorophyll to be the sole precursor of chlorophyll in the process of greening at higher temperatures, it would be necessary for it to be produced at an adequate rate. This possibility was tested by following the rates of development of protochlorophyll in the dark at 1° and 19° C. The barley leaves used for this experiment had previously been depleted of protochlorophyll by illumination at low temperatures at which no further chlorophyll was formed. Additional protochlorophyll was produced at 19° but not at 1°. This fact agrees qualitatively with the assumption that protochlorophyll is precursor to chlorophyll in the normal process of greening, since plants become green at 19° but not at 1° C. At 19° C., however, there is still some uncertainty as to whether the rate of formation of protochlorophyll is adequate to account for the rate of formation of chlorophyll.

By means of the techniques developed for the characterization and isolation of protochlorophyll, it has now been possible to demonstrate that protochlorophyll exists in barley leaves during the later stages of greening and also in mature leaves growing in the field. These observations are of particular interest because it is possible that chlorophyll acts in photosynthesis by a reversible dehydrogenation to protochlorophyll.

Mature green leaves contain a mixture of chlorophylls *a* and *b*. Usually the ratio of chlorophyll *a* to chlorophyll *b* is about 3 to 1. This ratio, however, changes greatly as chlorophyll is formed in etiolated leaves during illumination. Experiments with etiolated barley seedlings have shown that initially only chlorophyll *a* is produced. On continued illumination of the barley

seedlings, chlorophyll *b* appears and thereafter both chlorophylls increase. Gradually the usual ratio of approximately 3:1 for chlorophylls *a* to *b* is approached. The rates of formation of both chlorophylls are drastically reduced by lowering the temperature. Both rates are affected nearly to the same degree. Therefore, the two chlorophylls are produced in the same ratio at different temperatures. The results obtained so far make it appear improbable that chlorophyll *a* is the direct precursor of chlorophyll *b*.

The formation of chlorophyll in most of the higher plants is a photochemical process. In pine seedlings, however, chlorophyll is formed in the dark. By investigating chlorophyll formation in pine seedlings, factors not evident in the other plants may be disclosed which are of significance to the generalized interpretation of the process. *Pinus Coulteri* and *P. Jeffreyi*, furnished us through the generosity of Dr. P. Stockwell and Dr. N. Mirov of the Institute of Forest Genetics of the U. S. Forest Service, were used as experimental material.

Seeds of these species were germinated in the dark at 17° C. The cotyledons which emerged were dark green, although before germination they were white. The green cotyledons contained chlorophylls *a* and *b* in about the usual ratio: 3:1 in *P. Coulteri* and 3.7:1 in *P. Jeffreyi*. The presence of protochlorophyll was demonstrated in both species by chromatographic and spectroscopic examination, confirming older observations. Quantitative measurements showed, however, that only very small quantities of protochlorophyll were present, the ratio of chlorophyll *a* to protochlorophyll being about 50:1. Subsequent illumination of the seedlings had no significant effect on the quantities of chloro-

phyll *a* or *b* or of protochlorophyll in these cotyledons.

In contrast with the greenness of the cotyledons, the new growth of leaves and stems produced on 20-inch pine saplings in the dark is nearly colorless. The new needles produced when these saplings were placed in the dark contained traces of chlorophylls *a* and *b* and also protochlorophyll. They greened very slowly on illumination at room temperature. In the pine, then, there appear to be two different processes whereby chlorophyll is formed.

Chlorophyll formation varies in different genetic strains of plants. The examination of mutant strains possessing different capacities for the formation of pigments promises to yield valuable information concerning chlorophyll development. Various single-gene mutants in corn have been examined in regard to their ability to make protochlorophyll in the dark, to transform it to chlorophyll, and to accumulate sufficient chlorophyll to become green. These mutant strains were inbred for four generations at the University of Minnesota in collaboration with Professors E. C. Abbe, H. K. Hayes, and C. H. Burnham.

One strain of albino corn when germinated in the dark contained a normal quantity of protochlorophyll. Illumination of this strain at 17° C. for 5 minutes transformed protochlorophyll to chlorophyll, but further illumination for 6 hours almost completely destroyed the chlorophyll which had been formed. This particular albino mutant, therefore, possesses part of the mechanism for greening. Its inability to green arises from an excessive rate of destruction of chlorophyll in the light.

Several virescent mutants of corn, when germinated in the dark, produced only about one-tenth of the normal quantity of protochlorophyll. Illumination for 5 minutes transformed protochlorophyll to chlo-

rophyll. In contrast with the reaction in the albino, however, illumination for 6 hours produced additional chlorophyll. These virescent mutants are partially defective in their ability to make protochlorophyll, but possess the mechanism for the continued production and conservation of chlorophyll. The differentiation of steps in chlorophyll formation through genic influence is well illustrated by the use of chlorophyll-deficient corn mutants.

Inasmuch as illumination increases the chlorophyll content and photosynthetic capacity of dark-grown, etiolated seedlings, it is of interest to determine whether illumination increases at the same time the dye-reducing activity of the chloroplasts isolated from such seedlings. Barley leaves containing different quantities of chlorophyll were produced by exposing dark-grown seedlings to light for different lengths of time. Chloroplasts were isolated from the various lots of these seedlings and tested for their ability to photo-reduce dye. The greater the chlorophyll content, the greater was the dye-reducing activity of the chloroplasts. Plastid material obtained from unilluminated leaves had only a trace of activity. The conclusion is unmistakable that increase in chlorophyll content is accompanied by increased ability to reduce dye. Other factors affecting activity are also indicated.

SPECTROPHOTOMETRY

C. S. FRENCH, J. H. C. SMITH, H. C. PATTON, JR.,
AND B. G. RYLAND

An instrument has been designed for the automatic recording of the fluorescence spectra of leaves and algae. Since a need has also long been felt for an automatic recording instrument for absorption spectra, this equipment was designed to accomplish both these functions until a thoroughly satisfactory commercial instrument

for the recording of absorption spectra is available. Instruments now on the market do not cover the spectral range necessary for our purpose, nor do they use a narrow enough spectrum band. Much help on this problem has been obtained from Mr. D. Schuech, of the Department of Electrical Engineering, Stanford University. The basic components of the instrument are a constant light source, a grating monochromator, a photomultiplier tube with a light chopper and critically tuned amplification circuit, a correction device, and a commercial recording potentiometer. All necessary corrections are incorporated in a single curve which controls an attenuator through a photoelectric curve follower.

The instrument has now progressed to the point where performance tests are being made and the necessary modifications are being incorporated.

OCCURRENCE AND PROPERTIES OF CHLOROPLAST PIGMENTS

HAROLD H. STRAIN

In the long course of organic evolution, chloroplast pigments have played an indispensable role in the maintenance of living organisms. Within the plastids of living plants, these pigments utilize radiant energy for synthesis of organic matter and oxygen, both of which are required by plants as well as by animals. But the origin and the mechanism of this vital photosynthetic activity of the chloroplast pigments have yet to be discovered.

Clues to the evolution and functioning of the photosynthetic apparatus may be obtained from knowledge of the individual chloroplast pigments. These pigments have, therefore, been isolated from plants exposed to a variety of controlled conditions and from various plants growing under natural conditions. Considerable at-

tention has also been devoted to studies of the chemical properties of the pigments, particularly the reaction of the xanthophylls with acids.

The results of these studies, which supplement previous reports (Year Book No. 42 [1942-1943], p. 79; No. 46 [1946-1947], p. 90) and which are given in greater detail in the following sections, show that in a given plant, the pigments are remarkably constant in kind but somewhat variable in proportion. In different plants, the occurrence of particular pigments can usually be correlated with the phylogenetic or evolutionary pattern believed to have been followed by the organism. Here we have an independent confirmation of the evolutionary courses of plant development that were first deduced from taxonomic relationships based on morphological and reproductive characters.

The geological record shows that plants of the major taxonomic groups were present on the earth several hundred million years ago. Consequently, the chloroplast pigments must have remained constant through billions of generations while plants varied enormously in form and in reproductive habit. In view of the complexity and lability of the pigment molecules, especially their capacity for existence in various isomeric and stereoisomeric forms, this constancy of the pigments in the chloroplasts must be ranked among the great wonders of the organic world.

The constancy of the pigments in plants belonging to the principal taxonomic groups may present a limiting feature of the evolutionary processes of animals as well as of plants. For example, animals that require carotenoid pigments for vision and for vitamin supply may be limited in their variability by this constant mixture of pigments obtained from their basic plant food.

Exposure of live mature plants to light, to darkness, to oxygen, or to nitrogen for periods of several hours does not destroy the pigments or convert them into appreciable quantities of other colored substances. These observations indicate that photosynthesis is not accompanied by chemical changes of appreciable quantities of the chloroplast pigments; or if changes do occur, they must be quickly reversible. This result is in sharp contrast with the marked spectral changes that accompany the physiological action of hemoglobin and cytochrome in living organisms.

In killed plants, on the other hand, the chloroplast pigments are exceptionally labile, undergoing transformations into a variety of colored substances. These changes result from various reactions such as reversible and irreversible isomerization, induced oxidation, photochemical oxidation, and enzymatic hydrolysis.

Detection of the alteration of the pigments and isolation of the native pigments have required special attention to analytical methods, particularly to extraction of the pigments from the plant material and to resolution of the pigment mixture by the sensitive chromatographic adsorption technique. This experience has provided rules for use of the chromatographic adsorption method that have been useful in various studies of chemical compounds and their reactions.

Pigments of various plants. Pigments of over fifty species of higher plants have now been examined by chromatographic adsorption methods. The principal coloring matters are chlorophyll *a*, chlorophyll *b*, lutein, and β -carotene. Considerable quantities of the xanthophyll violaxanthin and very small quantities of the xanthophylls neoxanthin and zeaxanthin are always present. α -carotene often occurs in small proportions. Only in one plant was the

amount of zeaxanthin found to exceed the amount of lutein.

The pigments of several ferns and mosses and of a stonewort are identical with those of higher plants. Similarly, the chloroplast pigments of eleven species of cycads representing six genera are the same as those of higher plants except that small proportions of taraxanthin accompany the other xanthophylls. With a few exceptions, the pigments of green algae are likewise identical with those of higher plants, and they occur in about the same proportions. All these facts support the view that the photosynthetic systems of higher plants and of green algae evolved along similar pathways and then remained relatively constant during evolution of the numerous species known today.

Three fresh-water species of *Vaucheria*, a group of organisms commonly classified as green algae, yielded chlorophyll *a*, β -carotene, and a group of xanthophylls found in yellow-green algae such as *Tribo-nema*. This fact indicates that the *Vaucheria*s are yellow-green rather than green algae; hence further taxonomic investigation of this group is desirable.

A brackish-water organism described in last year's report as a *Vaucheria* may have been a siphonalean green alga. As this organism could not be found in a fruiting state, its taxonomic identity could not be determined with certainty; its pigments, however, were those of the siphonalean green algae rather than those of the fresh-water yellow-green algae. These results not only indicate the relationship of the organism, but they also show how information about the pigments may stimulate and supplement investigation in taxonomy and phylogeny.

The principal pigments from about sixty species of red algae have been found to be chlorophyll *a*, lutein and zeaxanthin,

β -carotene and α -carotene, and the red proteinaceous phycoerythrin. Many of these organisms contained little or no chlorophyll *d*. In one species, however, the chlorophyll *d* was equivalent to about 10 per cent of the chlorophyll *a*, in another to about 25 per cent.

In most red algae the principal carotenoids are β -carotene and lutein (a dihydroxy- α -carotene). But in species of the family Delesseriaceae, order Ceramiales, the principal carotenoids are α -carotene and lutein; and in species of the family Rhodomelaceae, order Ceramiales, the principal carotenoids are β -carotene and zeaxanthin (a dihydroxy- β -carotene). This variation in the proportions of α - and β -carotene and of lutein and zeaxanthin suggests that a complex physiological relation is involved in the formation of the several carotenoid pigments. This variation shows also that the possible role of these substances in photosynthesis can be played by either α - or β -carotene or by hydroxyl derivatives of either of these carotenes.

Effect of various conditions on the kind and proportion of the pigments. If photosynthesis were accompanied by chemical changes of the pigments, then different colored substances might be found when plants were exposed to intense light or to darkness. Thus far, no such change has been observed in mature plants. Several species of green algae exposed to sunlight for a day or to darkness either in air or in nitrogen for several days yielded the same pigments in the same proportions.

Some higher plants lose their pigments slowly when kept in the dark, but equivalent quantities of other colored substances are not produced. Other plants, as cycads, retain their pigments unaltered in the dark for several months. A few plants, such as certain green algae, synthesize their

normal complement of pigments in the dark. Only in a few instances, such as the winter reddening of foliage, is the loss of chlorophylls and carotenoids accompanied by the formation of appreciable quantities of other pigments. As in the ripening of fruits and in the development of flowers, these special pigments vary from species to species; their development is relatively slow; and there is no indication that they are associated directly with the photosynthetic process.

Action of acids on xanthophylls. When treated with acids, many xanthophylls are converted into other yellow substances some of which have been found in living organisms. Violaxanthin, obtained either from leaves or from pansy petals, yields, upon brief exposure to acids, a mixture of two pigments which can be separated by chromatographic adsorption and which exhibit spectral absorption maxima at wave lengths shorter than those of the maxima of the violaxanthin itself. These two pigments resemble the flavoxanthin of buttercups with respect to the wave lengths of their absorption maxima, but not with respect to other properties. Upon slightly longer reaction with acids, violaxanthin and the two flavoxanthin-like pigments yield other pale-yellow substances with absorption maxima at very short wave lengths and with other properties similar to those of xanthophylls that have been found in the eyes of chickens.

There are reports that violaxanthin of leaves yields with acids appreciable amounts of xanthophylls having a lower content of oxygen and with absorption maxima at longer wave lengths. This result may be explained as due to the loss of oxygen (or hydroxyl), an effect of possible significance in connection with the oxygen-evolution mechanism of photosynthesis. In all the experiments we have per-

formed thus far, however, violaxanthin has yielded primarily substances that absorb at shorter wave lengths. In the light of these results, it seems unlikely that violaxanthin splits off oxygen in the course of photosynthesis.

Chromatography. Chromatographic adsorption analysis is one of the most rapid and sensitive methods for the comparison of chemical substances. For example, if two substances suspected of being identical yield two bands in an adsorption column, the substances are considered to be different. Recent experience with leaf pigments shows that adsorption of a xanthophyll contaminated by a colorless impurity followed by adsorption of some of the pure pigment frequently leads to formation of two separate zones. This confusing effect can be eliminated to a large extent by mixing the two solutions before adsorption, or by reversing the order of the adsorption of the solutions.

It has also been found that certain impurities added to a pigment solution will cause the pigment to separate into two bands in an adsorption column. Some impurities may cause the band of adsorbed pigment to become very wide and diffuse; other impurities may cause the band to remain narrow with sharp boundaries. These effects point to new precautions to be observed in applications of the chromatographic adsorption method.

CHLORELLA AS A SOURCE OF FOOD

H. A. SPOEHR AND HAROLD W. MILNER

The liaison between the physical sciences and technology has become so intimate that discoveries and developments in these sciences find applications in industry with remarkable readiness. Such is not the case in the biological sciences, or certainly to a much lesser degree. The reasons for this slower application of advances

in the biological sciences are manifold, and at least to some extent reside in the great complexity of the biological industries and in the individualistic character of the human elements engaged therein, notably in agriculture. The growing of food for the earth's population is still in the hands of millions of independent-minded farmers; the plants they raise as crops were brought into cultivation by primitive man thousands of years ago. The increased production necessary to feed the constantly increasing population has been accomplished thus far largely through the introduction of machinery. From many sides serious question is raised whether these methods of food production are adequate to meet the demands of population growth. Food production, being based fundamentally upon the process of photosynthesis, is essentially a biological industry. It would seem to be, therefore, a compelling function of biological science to explore all possible means of contributing to the solution of this problem. It would appear, moreover, that biological science, in order to bridge the gap between discovery and application, may have to make a slightly greater effort toward application until the industries dependent upon it develop to the same point of awareness and acceptance of scientific research as is the case in the relations between technology and the physical sciences.

In so complex a problem as increasing the world's food production, involved as it is in innumerable climatic, nutritional, economic, and social complications, it would be temerarious to advocate a revolutionary change in methods of food production. Nevertheless, every effort must be made to improve existing methods and at the same time to explore all possible means which might supplement older, well established practices. In the course of such

exploration it would be almost providential if success were to come rapidly; it is rather to be expected that it can be attained only on the basis of a great deal of patient and painstaking search. Nor will it be advisable to place confidence in any single approach, or method, but instead numerous roads must be followed and any lead that seems promising should be pursued industriously and critically.

As a result of investigations conducted in this laboratory on the influence of environment on the chemical composition of plants, it was found that the percentages of fat, protein, and carbohydrate produced by the alga *Chlorella* can be modified within wide limits. Carbohydrates are relatively plentiful in the world supply; fats and proteins, on the other hand, are in deficit. Through the proper selection of culture conditions *Chlorella* can be made to produce about 50 per cent of its dry weight as protein, and under other conditions the same organism will produce as high as 75 per cent of its dry weight as fat. In fairly large-scale laboratory experiments such yields have been found to have a high degree of reproducibility, and some features of these investigations have been presented in previous reports (Year Book No. 42 [1942-1943], p. 83; No. 45 [1945-1946], p. 109; No. 46 [1946-1947], p. 93).

In the case of a process of this nature it can hardly be overemphasized that there exists an enormous gap between laboratory-scale experiment and large-scale production. In order to make evident the steps in the process of culturing *Chlorella* on a large scale which entail difficult scientific and engineering problems, some analysis has been made of these features. This has been done with a view to determining whether it would be justifiable to undertake the culture of *Chlorella* on what

may be termed a pre-pilot-plant scale. For this purpose there has been envisioned a plant of 300 to 500 gallons capacity of culture solution. A considerable body of experience is already extant on capacities somewhat smaller than this, and for various reasons of economy and availability of equipment, larger capacities than about 500 gallons would hardly be warranted.

An analysis of this kind naturally includes much detail, so that only the outline of the types of problems can here be given. There is, in the first place, the question as to whether a batch method should be adopted or whether a continuous process can advantageously and safely be undertaken. This question in itself has a number of ramifications involving the adoption of reliable methods of sterilization and especially the all-important factor of obtaining the maximum amount of illumination in the culture containers. This latter problem is in many respects the most crucial one, for on it depends the photosynthetic activity of the organism and the production of all the organic matter which is elaborated. Consequently, the success of the process, in terms of yield and the chemical nature of the organic material formed, depends in a large measure on the architecture of the culture containers.

As a result of experience gained from laboratory-scale experiments it is clear that natural illumination, under favorable climatic conditions, offers many advantages over artificial illumination for a large-scale undertaking, even aside from the question of cost. This conclusion is based upon experience with fluorescent as well as incandescent forms of artificial light. On the other hand, with the use of sunlight consideration must be given the question of the infrared radiation. The cells grow best at about 20° C.; under high illumination, and with the proper nutrient media, they

will stand temperatures as high as 40° for a short time without any apparent deleterious effects, but not higher. Under some circumstances, therefore, the use of natural illumination demands appropriate shading from direct insolation, or filtration of the infrared rays. The question of the most favorable geographical location for a large-scale undertaking is also of considerable significance, and some study has been given this matter. In some regions a production plant could operate effectively for the entire year, whereas in other areas this would be possible for only one-third of this period. The cells of *Chlorella* will grow under a wide range of environmental conditions; their yield and composition vary greatly, however, especially in relation to the amount of light energy they receive. It need hardly be stressed that composition is revealed only through chemical analysis.

There are many questions in connection with the harvesting, handling, drying, and storing of the *Chlorella* cells which will require careful study. The cells respire material which is stored in them, and even after drying, some of the components are subject to change and deterioration.

It is impossible to make unqualified statements concerning yields. These, in terms of the weight of cells or of product desired, for example protein or fat, depend upon a number of factors. Most important among these factors are the following: (1) the composition of the nutrient medium, (2) the length of time the cultures are grown, and (3) the intensity of illumination. The production of fat may serve as the best illustration. A fat content of 55 per cent of the dry cells would represent a conservative figure, the maximum thus far attained having been considerably higher. On the basis of 55 per cent fat, to produce 1 pound of fat would

require 175 gallons of culture solution isolated with high light intensity for 80 days. Under favorable conditions and in a suitable locality, 3 to 4 pounds of fat might be produced from 175 gallons of culture solution in a year. Higher yields of fat can probably be produced, and it is also conceivable that somewhat greater rates of growth can be attained, but on the basis of present information it would probably not be warranted to expect more than 5 pounds of fat from a volume of 175 gallons of culture solution per year.

As has already been stated, the shape or configuration of the culture chamber is of great significance for the development of the algae. The most important requirement is that a relatively large surface be exposed to light, while at the same time provision is made for adequate sterilization, aeration, and easy filling and emptying of the chamber. The batch or unit process has several advantages. If glass cylindrical containers 7 inches in diameter and 40 inches tall, with a capacity of 6 gallons each, were used, 29 of these would be required for the 175 gallons of culture medium already referred to. On the other hand, if the container were in the form of a glass pipe 7 inches in diameter, the 175 gallons of medium would require 87 linear feet of such pipe. A more desirable dimension for illumination purposes would probably be a pipe of 4 inches diameter, and this would require 268 linear feet for the same volume of liquid. Finally, a culture chamber in the form of a flat tank, with the culture solution 3 inches deep, would require an area about 10 feet square. These examples will suffice to give an approximate indication of the culture chamber requirements for the production of 1 pound of fat in a growing period of about 3 months. They represent conservative estimates. The formation of fat is a

slow process and requires the fixation of more energy than does the production of either carbohydrates or proteins.

It is exceedingly difficult to make a sound comparison between the production of fat, for example, by the culture of *Chlorella* with its production by means of agricultural crops. In the case of *Chlorella* culture, only the insolated area of the container of the culture solution is of significance. On the basis of the area of the illuminated portion of the culture vessels used, the yields of fat from *Chlorella* are compared in table 2 with yields of fat from farm crops. The farm crop data are taken from *Agricultural statistics 1946* (U. S. Department of Agriculture) and are the highest reported for each crop. The figures for fat content of the oil-producing farm crops are taken from G. F. Jamieson's *Vegetable fats and oils* (1943). The yields are calculated on the basis of pounds of fat per acre per 120 days, representing a crop year. For a *Chlorella* culture undertaking to justify itself, it would certainly have to be run for the greater part of the year, which so far as the organism is concerned is perfectly feasible; and the annual production could be correspondingly increased. Most crop plants, on the other hand, are confined to certain seasons or limited portions of the year, which means, of course, that for most of the year the solar energy falling on the land is not utilized for the production of organic carbon.

The values given in the table cannot be interpreted too rigidly; they are given for general comparative purposes only and in order to give some idea of the productive capacity of *Chlorella* cultures for fat, one of the most difficult of food constituents to obtain. The production of *Chlorella* has been carried out on a laboratory scale; methods for large-scale production, even

of pilot plant size, have not been developed. It appears that a commercial venture would be economically hazardous.

One further matter is worthy of mention, and this pertains to the nature of the products synthesized by *Chlorella*. The chemical nature of the fatty acids has been thoroughly investigated. These acids

TABLE 2

PRODUCTION OF FAT BY VARIOUS CROP PLANTS
(FROM FARM CROP DATA) AND BY CHLORELLA
(CALCULATED FROM SMALL-SCALE LABORATORY
EXPERIMENTS)

Source	Tons per acre per crop year, dry wt.	Per cent fat	Pounds fat per acre per growing season of 120 days
Chlorella.....	2.85	7.0	400
	1.74	17.0	593
	1.66	42.0	1390
	1.15	56.5	1303
	0.71	55.0	747
	0.46	75.0	687
Maize (grain).....	1.37	4.5	123
Flax (seed).....	0.70	37.0	518
Soy bean (seed)....	0.63	18.0	227
Peanuts (nuts).....	0.60	30.0	360
Cottonseed.....	0.14	20.0	56

are for the most part highly unsaturated, which would indicate that the fats of which they are a part would be easily assimilated. On the other hand, there is evidence that the fats are not easily extracted from the intact cells, nor is it known whether the fats are available if the cells are fed to an animal organism. Little is known of the nature of the proteins of *Chlorella*, of the nature of their constituent amino acids, and as to whether the protein in the cells can serve a higher animal as food. These matters are here mentioned as examples of the type of problems which are still to be solved in connection with the use of *Chlorella* as a source of food.

THE URONIDES OF LEAVES

H. A. SPOHR

For many years it has been assumed that the first product, and probably the only direct one, of the photosynthetic reaction occurring in plants is a compound of the general composition of a simple sugar. Much of the theoretical work on the mechanism of photosynthesis has been based upon this fundamental assumption. Although there appears to be considerable experimental evidence to support it, more recent investigations, including those carried out with radioactive carbon, indicate that this assumption may not be so universally applicable as was believed. Evidence is growing that the photosynthetic reaction is more intimately linked with the general metabolism of the plant than has been realized, and that the course of synthesis may follow different directions and result in a variety of products. This concept is as yet very much in a state of flux, and no accurate generalizations are possible. Yet it is important to realize that although the plant under most conditions synthesizes primarily carbohydrate, the possibility of the synthesis of other types of compounds does not seem to be excluded, and this fact must be considered in the formulation of any theory of the mechanism of the photosynthetic reactions.

A great deal of study has already been devoted to the chemistry of carbohydrates in connection with their formation in photosynthesis, and to the subsequent changes which they undergo in the plant. To a lesser extent, fats and proteins also have been investigated from this point of view. The chemistry of the members of these important groups of naturally occurring compounds has been carried to a high level of exactness. As constituents of leaves there are, however, several groups of organic compounds which are still only imperfectly

known and whose possible relation to the photosynthetic process has not been adequately considered. One of the most striking deficiencies in our knowledge of leaf constituents is that concerning the group of uronides, which are substances in which uronic acids are combined with various carbohydrates into very complex molecules.

The investigations which have thus far been carried out on the composition and properties of uronides have for the most part used as a source the structural elements of plants, such as wood, storage organs, exudates, and mucilages. In animal tissue also the so-called hyaluronic acid plays an important role in holding cells together in a jelly-like matrix. During the past year uronides have been isolated from the leaves of plants, and efforts have been made to purify these preparations. Fractions have been obtained which contain about 30 per cent uronic acid, the rest being chiefly carbohydrate. With greater purification and increased uronic acid content, the preparations become increasingly soluble in water and are precipitated with difficulty from water solutions. The free uronic acids are exceedingly labile, and their purification and analysis present many difficulties, so that only a beginning has been made on this problem.

In an effort to simplify the process of hydrolysis of uronides and polyuronic acids, a method has been devised which makes use of 90 per cent formic acid as a hydrolyzing agent. This method of hydrolysis, in certain cases, has some advantages over older methods in that it obviates the necessity of removing the acid used for hydrolysis through neutralization, and also because formic acid does not cause decarboxylation of the uronic acids. By the use of this method alginic acid, a polymannuronic acid, has been hydrolyzed with a yield of about 45 per cent d-man-

nuronic acid lactone. By the same means pure crystalline products, in satisfactory yield, have been obtained by the hydrolysis of starch, gum arabic, ivory nuts, and

similar polysaccharides, but it has not been possible by this method to attain in a satisfactory manner the hydrolysis of pectic acid.

EXPERIMENTAL TAXONOMY

JENS CLAUSEN, DAVID D. KECK, AND WILLIAM M. HIESEY

Progress during the past year has been made chiefly in two lines of investigation. One has been the continued advancement of the range-grass program, which was described in some detail in the last Year Book. The other has been the analysis of a long-term study on the ecological genetics of climatic races of *Potentilla glandulosa* which was begun in 1932 and is now sufficiently matured to bring to publication. In addition to these major activities, preliminary study of promising future lines of investigation has been under way.

THE RANGE-GRASS PROGRAM

Some 80 new second- and third-generation progenies of *Poa* containing potentially useful economic strains, and consisting of some 10,000 seedlings, were started in the greenhouse in January 1948, but the drought in California during the early part of the season made it inadvisable to plant them in the field during the spring as originally planned. They are being held over in lathhouses, and field planting will be made in the fall with the coming of the first winter rains. From these seedlings, however, it is already evident that asexually reproducing forms may arise in the third generation from sexual hybrids that originated as offspring of partially asexual species. This makes possible the selection of new self-perpetuating strains from widely segregating hybrid populations, and greatly increases the possibilities of finding new desirable strains.

The active co-operation of the U. S. Soil Conservation Service in the *Poa* pro-

gram has greatly enhanced the scope of the work. In particular, the testing of hybrid progenies in various climates, especially in the nurseries at Pullman, Washington, and at San Fernando, California, has already provided important information concerning the possibilities and limitations of various hybrid lines. The importance of studying the climatic fitness of derived lines is demonstrated in spectacular fashion by strains that thrive under one climate but fail in another.

The Carnegie Institution staff has made comparative studies of the grass hybrid plantings at the Soil Conservation Service nurseries and at its own stations at Stanford, Mather, and Timberline. The extensive plantings at Pullman received detailed attention from Drs. Clausen and Keck, who were joined by Dr. A. L. Hafenrichter, head of the nursery division of the Service; and the smaller planting at San Fernando was studied by Clausen. In response to an invitation extended by Dr. Etlar L. Nielsen, of the Division of Forage Crops and Diseases of the Bureau of Plant Industry, Clausen and Keck conferred with him on problems arising from his extensive cultures of *Poa pratensis* on the University of Wisconsin campus. During a 5-day field trip through the North Coast Ranges of California, Mr. Harold W. Miller, manager of the Pleasanton Nursery of the Soil Conservation Service, and Dr. Keck collected new strains of *Melica* and other grasses.

On request from the Ministry of Agriculture at Amman, Transjordan, a sample

of some 19 hybrids was sent to Amman to be used in a program for improving range grasses for the winter-rain areas in this east Mediterranean plateau region.

CLIMATIC RACES OF *POTENTILLA GLANDULOSA*

It is a general evolutionary principle that the ability of a species to occupy different environments depends upon its ability to evolve races especially adapted for living in them. The fitness of these races to the environment is largely physiological, but the physiology is controlled by genes. The analysis of the genetic structure of climatic races is therefore of primary importance for the understanding of evolution.

Potentilla glandulosa, of the rose family, is a species well adapted for such investigations. It is highly variable and contains eleven or twelve morphologically distinct subspecies in western North America. Four of these occur in the climatically very diverse transect across central California, where they have different geographic ranges. Morphologically they are as distinct from one another as many taxonomic species, yet they are completely interfertile. Natural hybrids occur where the subspecies meet, but these constitute only a minor part of the total natural population of the species. All forms of this species and its close relatives have only 7 pairs of chromosomes.

In the Coast Ranges of California grows subsp. *typica*, a form of compact habit with large, coarse leaves and small, whitish, erect petals nearly hidden by the large sepals. It is active the year round, but makes its principal growth during the winter and early spring. It contains at least two ecotypes: one in the Outer, and another in the Inner Coast Range.

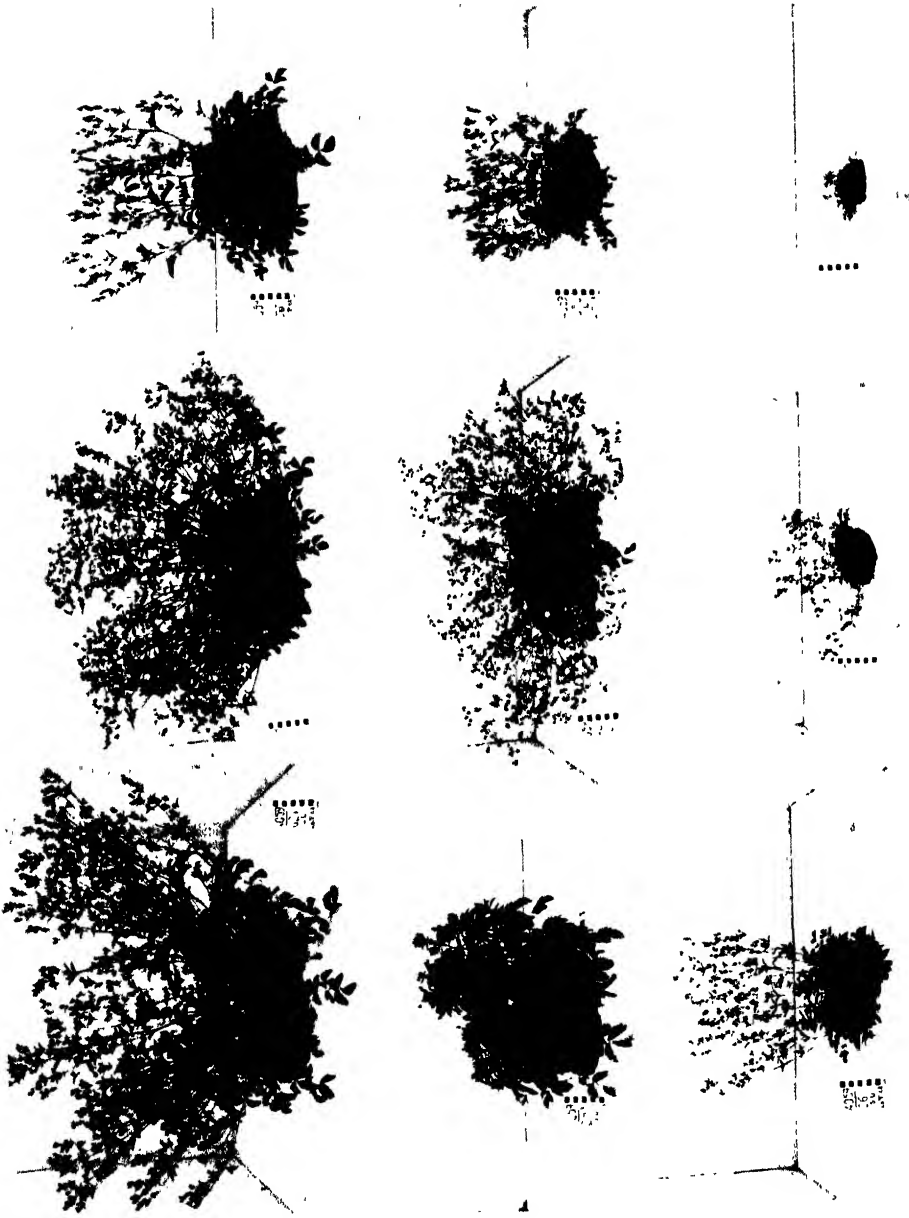
In the foothills of the Sierra Nevada subsp. *reflexa* occurs at altitudes between 800 and 6000 feet. This has an openly

branched habit and small, yellow, reflexed petals. At least two ecotypes exist within this subspecies. One is a form from the dry oak savannas below 2000 feet, and the other grows on slopes within the coniferous forest belt at higher altitudes in a region of greater rainfall. The former is winter-active and early flowering and the latter winter-dormant and late when grown in the mild lowland climate at Stanford.

In the upper half of the range of *reflexa* another subspecies, *Hanseni*, is found, but the two subspecies are in different local habitats. *Hanseni* grows in moist, cool meadows at from 4000 to 8500 feet altitude, whereas *reflexa* occurs on the warmer slopes. The two differ morphologically, *Hanseni* being of much stricter habit and having large, white, upturned petals that contrast with the small, yellow, recurved petals of *reflexa*. Subspecies *Hanseni* also contains at least two ecotypes, for the forms from lower altitudes bloom about two weeks later than those from higher altitudes when grown in the same garden.

At altitudes from 8000 to 11,000 feet still another subspecies can be recognized. This is *nevadensis*, which is dwarfer than *Hanseni*, with small leaves, slender rhizomes, and large, whitish, spreading petals. Within *nevadensis* the relatively tall subalpines from 8000 to 10,000 feet belong to a different ecotype from the dwarf alpine from 11,000 feet that remain dormant longer but develop faster in spring.

The climatic tolerances of all these races were previously tested by growing cloned representatives in the transplant gardens (see Year Book No. 37 [1937-1938], p. 218). In these experiments it was found that plants of subsp. *typica* from the coastal region are vigorous at the lowland station but somewhat weaker at the mid-altitude one, and they seldom are able to survive even the first winter at the alpine station. Likewise, all forms of the foothill sub-



Examples of the variation among second-generation offspring of a hybrid between a coastal race (subsp. *typica*) and an alpine race (subsp. *nevadensis*) of *Potentilla glandulosa*. All were growing in a uniform garden at Stanford.

species, *reflexa*, die promptly at Timberline, but they thrive best at mid-altitudes. Plants of the mid-altitude meadow subspecies, *Hanseni*, survive at all three stations, but grow by far the best near their native home at the Mather station, and are much weaker at Stanford. At Timberline, they are also weak and they flower too late to produce ripe seed except in the most favorable years. The subspecies *nevadensis* is the most vigorous and frost-resistant form in the high-altitude garden, and the only one able to produce seed there regularly. These four subspecies, therefore, differ as much in their ranges of tolerance of environmental conditions as in their morphology, and each one is well fitted to its particular natural environment.

GENETIC ANALYSIS OF THE CLIMATIC RACES

Hybrids between representatives of the Coast Range, Sierran foothill, and alpine races of the subspecies *typica*, *reflexa*, and *nevadensis*, respectively, were highly fertile. Second-generation progenies of only two of these combinations were studied in detail. One, of 1000 individuals, was of a cross between *typica* from the mild southern California coast near Santa Barbara and *nevadensis* from the rigorous alpine climate near Upper Monarch Lake at 11,000 feet altitude in the southern Sierra Nevada. The other, of 578 individuals, was obtained from a cross between *reflexa* from the warm Sierran foothills of Tulare County at 2500 feet and a race of *nevadensis* from slopes near the Timberline transplant station at 10,000 feet altitude. Every plant of the latter F_2 was cloned and grown for a period of from seven to nine years at the three transplant stations to test its responses to the lowland, mid-altitude, and alpine environments. The accumulated data were averaged to

give as dependable a picture as possible of the performance of each individual. Furthermore, 20 F_3 progenies from selected F_2 individuals of this cross were grown at Stanford to facilitate further genetic analysis. The vigor of the hybrids was maintained through both the second and third generations, demonstrating that these climatic races are conspecific, although they are as distinct morphologically as the average species.

The segregation of the hybrids in the two F_2 populations was striking, with scarcely two plants alike. Examples of some of the segregating types are shown in plate 1. In one F_2 population 14 different characters were tabulated for each individual plant, and in the other, 15. Each character was observed to vary by a series of small steps from one parental type to the other. The steps were too gradual to score with complete certainty, but they were somewhat more easily recognized in the F_3 . The characters studied included height of plant, length of leaves, number of stems, amounts of pubescence and anthocyanin, height of root crown below the basal rosette of leaves, density of inflorescence, size of sepals, length, width, and shape of petals, their orientation and color, and weight and color of seeds. In addition to these characters, physiological differences were also scored, such as winter activity as compared with winter dormancy, earliness of flowering, frost resistance, and the modifications in growth at the three stations.

The systematic study of these segregating characters posed a difficult problem in genetic analysis. This task, however, was greatly facilitated by the use of punch cards and sorting machines. The expression of each character studied was numerically graded, the value of the alpine parent being arbitrarily set at 1, and that of the lowland parent at a higher number, de-

pending on the number of classes into which the plants were graded with respect to a given character. The number of classes ranged from 2 to 10. The character expressions of each plant were coded and the code was transferred to punch cards. The cards were then systematically machine sorted, so that the frequencies of the character associations in all possible combinations could be counted and used in computing correlation coefficients. The punching and sorting of the cards was done under contract at the San Jose plant of the International Business Machines Corporation. The statistical work, especially the computation of the correlation coefficients, was done mostly by Dr. Helen Sharsmith.

Analysis of the data made it possible to estimate the probable number of genes determining the characters distinguishing the alpine and the lowland races. For the characters studied, this estimate ranged from 2 to 5 pairs of genes. None of the characters was governed by a single gene. The presence of predominantly multiple genes is demonstrated with particular clarity in the diploid *Potentilla glandulosa*, with only 7 pairs of chromosomes. No basic difference was found between the mode of inheritance of morphological characters, such as petal shape, and that of physiological characters, such as winter dormancy. It is estimated that, as a minimum, some 60 pairs of genes are responsible for the differences investigated; these, of course, do not include all the character differences that are known to segregate.

In some cases, such as leaf length, the character appeared to be determined by three or four genes each with equal effect; in others, one gene in the series appeared to outweigh the others, which then functioned as modifiers. Commonly a combination of genes with a positive effect was counterbalanced by inhibitors, resulting

in segregations in ratios of 13:3 and 61:3. The orientation of petals, for example, was determined by one pair of genes that reflex them unless either one of two pairs of genes for upcurved or erect petals is present. Color of petals was likewise determined by a series of genes that affect the intensity of the basic yellow pigment, and these are counterbalanced by another series of genes with a bleaching to whitening effect. Even in the F_2 's the segregation for petal color was found to be complex. It is estimated that approximately five pairs of genes are responsible for the differences in petal color between the foothill and alpine races—two pairs determining the intensity of yellow, one pair for bleaching the yellow to cream or creamy yellow, and two pairs of whitening genes which, when present, mask the effect of the others.

Linkage was found to exist between physiological and morphological characters. It is significant that in almost every instance of linkage the character pairs were found to be associated in the same combination in which they occur in the parental types. Linkage was far from absolute, however, for recombinations of all kinds of characters, whether morphological or physiological, occurred with considerable frequency.

SELECTION EXPERIMENT

The F_2 population consisting of 578 individuals that were individually cloned and grown simultaneously in the contrasting climates of the three transplant stations has been under observation since 1938 (see Year Book No. 37 [1937-1938], p. 221; No. 39 [1939-1940], pp. 162-163). The gardens in which it has grown serve as a series of screens that select forms adapted to specific kinds of climates.

In the three environments, the growth

patterns of the segregating F_2 clones of this cross between a form of *reflexa* from the Sierran foothills and an alpine form of *nevadensis* were actually more diverse than those found by testing wild forms of *Potentilla glandulosa* from many different climates. These numerous growth patterns recombined in various ways with morphological characteristics inherited from the grandparents, resulting in individuals that had the appearance of alpiners but were successful at low altitudes, and vice versa, as well as many other types. This is in marked contrast with the native foothill and subalpine parental forms, which have well marked characteristics and were found to be definitely best fitted to lower and higher altitudes, respectively.

Among the segregating F_2 plants were "super-alpiners" that thrived much the best at the alpine station, for, unlike the native forms from this vicinity, they did not gain in growth when transplanted to the milder climate at Mather. In this respect they resemble certain strictly alpine species that occur only on the high crests of the Sierra Nevada. Some of the synthetic "super-alpiners" exceeded in vigor the native forms at Timberline, and many at the same time possessed morphological traits of the foothill parent, such as a relatively large size.

Another group of segregants in this F_2 reacted like forms native to the coast in that they grew most vigorously at Stanford, but they differed from true coastal natives in being winter-dormant at this lowland station. Some individuals of this class were successful only at the two lower altitudes, but others survived at all three stations although weakly at the alpine station.

In addition to these contrasting classes, some F_2 plants had such a wide range of physiological tolerance that they grew successfully in all three environments. They differ in this respect from any forms

found in the wild, for in all trials with the latter, the plants show better survival and growth at one of the stations than at another. Probably special physiological balances between the temperature coefficients of the assimilatory and respiratory processes are required to permit such individuals to grow successfully in environments so different.

It is evident that selection for many different environments is possible from the diverse biotypes composing such an F_2 population. The intraspecific interchange of genes between races from contrasting climates releases new evolutionary possibilities and makes possible the building up of forms fitted for new climates, as well as of forms that are more tolerant to wide differences in climate than are those already existing. Such crossings seldom occur in the wild because races from such contrasting climates rarely, if ever, occur together.

The differences between climatic races are governed by an intricate genetic system. Many character differences distinguish the races, and each character is determined by a series of multiple genes. In *Potentilla glandulosa*, with only 7 pairs of chromosomes to carry the genes, essentially all the characters are genetically linked together through one or more of their determinative genes. This situation tends to preserve the genic diversity in the species even under radical changes in the environment. On the other hand, through the mechanism of crossing-over and recombination, the system is flexible enough to permit the production of new forms fitted to new environments. Racial differences are determined by series of genes, each with a minor effect, rather than by a single pair of genes with a major effect, and this tends to reduce any deleterious physiological effects that may be caused by an individual gene.

These studies in *Potentilla* have shown

that the races of one species may store a great potential of genetic variation from which new races can be built, and that genetically there are no differences between physiological and morphological characteristics, although it is essentially the former rather than the latter that determine the survival of the plant in its particular environment.

EXPLORATORY CROSSINGS

The crossing of contrasting latitudinal and altitudinal races of *Achillea borealis* and *A. lanulosa* of the *millefolium* complex is being attempted this year with the object of studying their genetic-physiological structure. New strains from Iceland, kindly supplied by Dr. Steindor Steindorsson, and forms from the highlands of Mexico, collected by Mr. Oliver Norvell, are being included in the program of crossing. *Achillea*, being a composite with flowers difficult to emasculate, and forming a polyploid complex with

relatively high chromosome numbers, is not so well suited technically to genetic experiments as *Potentilla glandulosa*; however, the more complete array of climatic races in *Achillea* and the ease with which it may be propagated vegetatively make the group better adapted to physiological experiments.

Preliminary crossings are also being attempted within the *Mimulus guttatus-Tilingi* group of the Scrophulariaceae as part of a search by different laboratories on the west coast for a group of plants having as nearly as possible ideal characteristics for co-operative investigations on genetics, cytology, physiology, ecology, and evolution, as outlined last year at the informal conference at Mather (see Year Book No. 46 [1946-1947], pp. 103-104). This group of *Mimulus* appears to have characteristics that favor rapid genetic investigation, and is of ecological interest because of the apparent existence of numerous climatic races.

PALEOBOTANY

RALPH W. CHANEY

Fossil representatives of the coast redwood, *Sequoia sempervirens*, a tree now native only on the coast of California and Oregon, have been widely recognized by paleobotanists and botanists since Heer first described *Sequoia langsdorfii* from the Miocene of Switzerland in 1855. Cretaceous and Tertiary species of *Sequoia* have been well known in the United States since the pioneer work of Lesquereux in the '70's. Many impressions of leafy twigs and cones referred to this genus have been found not only at middle latitudes, where the coast redwood now lives, but in Greenland, Spitzbergen, Siberia, Alaska, and the arctic islands of North America, in some instances far beyond the present northern limits of trees of any kind.

There are questions involving this pattern of occurrence in past ages which have been difficult to answer. How could a tree now confined to the mild, moist borders of the Pacific have lived in the John Day Basin, where present-day climate shows wide extremes of temperature and scant rainfall? Major changes in climate, partly involving the late Tertiary upbuilding of the Cascade Range, have been invoked in explanation. How could a tree which cannot now survive cold winters have ranged northward to Grinnell Land, within eight degrees of the North Pole? This has been explained on the assumption not only of climatic changes in later geologic time, with polar ice caps and climatic extremes considered a geographically late

development, but by the suggestion that the northern redwoods of the past may have been hardy ecotypes which shed their leaves during the long winters of high latitudes.

Another sort of question has arisen in connection with the occurrence in Oregon and elsewhere of *Sequoia* cones attached to long naked stems. Lesquereux assigned distinct specific status to cones of this sort, and for the past decade or more there has been speculation on both sides of the Pacific regarding their relationship to cones of other species of *Sequoia*, fossil and living, which are borne on leafy twigs.

This latter question was solved in 1941 by the Japanese paleobotanist Miki, who pointed out that the long-stemmed fossil cones have oppositely arranged scales, whereas those of *Sequoia* are spirally attached; he further noted the occurrence of these cones in the same deposits with leafy twigs on which needles occur in opposite position along the twig, again in contrast with their spiral arrangement in *Sequoia*. He suggested that these leafy twigs were deciduous, like those of certain other members of the family Taxodiaceae, but unlike those of *Sequoia*, which is evergreen. A new genus, *Metasequoia*, was founded to include two species from the Tertiary of Japan. Miki's paper, issued during the war, had probably not been read by anyone outside of Asia when the announcement was made, during the spring of 1946, that three living trees of a kind never before seen had been discovered during the preceding year in Szechuan. Specimens brought in by a forester named Wang were studied by Professor W. C. Cheng, of National Central University, and Dr. H. H. Hu, of Fan Memorial Institute of Biology, who reached the brilliant conclusion that these trees were living representatives of the genus *Metasequoia*. Further field work, supported in

part by the Arnold Arboretum of Harvard University, resulted in the discovery of several hundred trees to the south, in western Hupeh. Hu and Cheng have recently described this new species as *Metasequoia glyptostroboides*; it is characterized by long-stemmed, decussate cones, and by opposite leafy twigs with oppositely arranged needles, like those of the fossils described by Miki; the male flowers, borne in long panicles, resemble those of the swamp cypress, *Taxodium*, but differ in having the typical opposite arrangement characterizing other organs of *Metasequoia*. Hu also determined that certain fossil species which had been assigned to *Sequoia* had the characters found in *Metasequoia*, and published a short paper assigning fossil material from Manchuria to this new genus.

A survey of the specimens assigned to *Sequoia* from rocks of Tertiary and Cretaceous age in North America is revealing the fact that many of them also have the cone and foliage characters of *Metasequoia*. This is particularly true of specimens from high northern latitudes, on this continent and elsewhere. By contrast, material described as *Sequoia* from Europe appears to be properly referable to that genus, and no specimens referable to *Metasequoia* have as yet been noted there. A preliminary survey of the fossil record from the mainland of Asia indicates the absence there of *Sequoia* during Cretaceous and Tertiary time, and the widespread occurrence of *Metasequoia*. At middle latitudes in North America, both *Sequoia* and *Metasequoia* are recorded in rocks from Cretaceous to Miocene in age; *Metasequoia* appears to have become extinct thereafter, and *Sequoia* has been restricted in distribution. In Asia the range of *Metasequoia* has likewise become progressively more limited, until it is now found living only

in the deep valleys of western Szechuan and adjacent Hupeh, in Central China.

For the purpose of studying the modern environment and associates of *Metasequoia*, Dr. Chaney visited this region in March 1948. A few large trees and several hundred small ones were seen growing on moist slopes and valley bottoms at altitudes from 4000 to 4500 feet, under conditions which appear to be essentially natural. Associated trees include chestnut (*Castanea*), sweetgum (*Liquidambar*), a small-leaved oak (*Quercus*), birch (*Betula*), beech (*Fagus*), cherry (*Prunus*), and katsura (*Cercidiphyllum*), all of which are deciduous. This group of trees is widely distributed at middle altitudes in Central China, and constitutes a well defined plant formation which has been recognized by Cheng and others. It is this assemblage of genera, together with others not as yet seen living in immediate association with *Metasequoia*, that is widely recorded at middle latitudes in North America and Asia during later Tertiary time, and that ranged widely around higher latitudes during early Tertiary time. The southward migration of this forest from high to middle latitudes during the Tertiary period, which has been previously described for *Sequoia*, is thus established; the prevailingly deciduous habit of all its members, including *Metasequoia*, is consistent with the northern origin which the fossil records indicate. Survival of this unit of vegetation down to the present, over a span of geologic time to be measured in scores of millions of years, has been made possible by the perpetuation of Tertiary climate, at least in approximate measure, in the sheltered interior of Asia. This region therefore becomes the key to our understanding and interpretation of many of the fossil floras of the northern hemisphere. The forest which lives there is

perhaps the most notable case of group continuity through time which is known anywhere in the world.

The pressure of overpopulation has destroyed most of the natural vegetation of China, and even in the remote interior provinces where *Metasequoia* has been found there remain no extensive forests. It is a matter of only a few years before the wooded ravines in which this tree has taken refuge will be cleared for agriculture; its wood is already being used for fuel and lumber. Recent formation of a committee in China whose purpose will be to protect some of the stands of *Metasequoia* and its associates is an encouraging development on a continent where conservation has until now been given little emphasis.

In view of the uncertain status of fossil material previously assigned to *Sequoia*, the study of the fossil floras of the John Day Basin has been temporarily set aside. Assignment to *Metasequoia* of most or all of the specimens formerly considered to represent *Sequoia langsdorffii* and *Sequoia heerii* in the Bridge Creek flora, of Oligocene age, and of many of the specimens assigned to these species from the Miocene Mascall flora, gives the middle Tertiary floras from eastern Oregon an interior facies which is wholly consistent with their present geographic position. By contrast, the Rujada flora, of approximately the same age, but located nearer the coast line of the middle Tertiary, is not known to include *Metasequoia*; a true *Sequoia*, probably the ancestor of the living coast redwood, is the common conifer of this coastal forest of the past.

Not all the questions regarding *Metasequoia* and *Sequoia* have yet been answered. We do not know why *Metasequoia* became extinct in North America some fifteen million years ago; the presence

of fossil *Sequoia* at some interior stations like Florissant, Colorado, and Yellowstone Park during the earlier half of the Tertiary is not consistent with its present coastal habitat; not nearly all the necessary information on the occurrence and associates of the living trees of *Metasequoia* is as yet available, although current studies by

Chinese botanists are adding materially to our knowledge. But we are at least, and at last, in a position to distinguish these two related genera when we find them in the rocks, and to reconstruct their habitats by comparison with the environments of their now widely separated living equivalents.

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DEPARTMENT OF EMBRYOLOGY

Baltimore, Maryland

GEORGE W. CORNER, *Director*

The work of the Department of Embryology continued during the year on an even course. There were no serious distractions, and only one change was made in the permanent research staff. This was the appointment, as of December 1, 1947, of Dr. David B. Tyler, who came to us, after an interlude in military research, from the California Institute of Technology, where he was Research Fellow. Dr. Tyler's experience has largely been in tissue metabolism, especially of the carbohydrates, and in his work with us he is applying the methods of research in that field to problems of embryonic metabolism. Dr. Tyler is working in close association with Dr. Flexner.

Dr. Irwin H. Kaiser, who worked with us during the year 1946-1947, and who held the rank of Fellow in the Department during the second half of that period, left June 30, 1947 to join the resident staff in obstetrics at Sinai Hospital, Baltimore.

Dr. E. Carl Sensenig, now Associate Professor of Anatomy in the University of Alabama, returned in the summer of 1947 as in several previous years, to continue his work on the embryonic development of the human spinal column.

Dr. Charles M. Goss, professor of anatomy at Louisiana State University, spent about two weeks with us in March 1948, for the purpose of studying the earliest stages of the heart and blood cells in human embryos. Dr. Goss has made fundamental observations on the heart in living mammalian embryos, especially of the rat, and wished to apply to the human embryo what he has learned about the time of the first heartbeat and the beginning of the blood circulation. Com-

ments on the result of his work will be made below, under "Program of investigations."

Dr. Alexander Barry, Instructor in Anatomy, University of Michigan, spent a week working with Dr. Reynolds in 1947. This time was devoted to working out the application of the technique of measuring pressure in blood vessels, etc., with the highly sensitive strain-gauge manometer system which Dr. Reynolds has constructed. As an extreme test of the method, Dr. Barry and Dr. Reynolds were able to get a clear graphic record of the intracardiac pressure from the beating heart of a 72-hour chick embryo.

By action of the Institution in October 1947, Dr. Joseph Gillman, of the University of the Witwatersrand, Johannesburg, South Africa, was appointed Research Associate of the Department of Embryology for the year 1948. This action recognizes the contribution made by Dr. Gillman in his monograph on the development of the human ovary and testis, summarized below, and his co-operation in obtaining early embryos of the baboon.

Less than a month after the end of the year covered by this report, Dr. George L. Streeter, Director of the Department of Embryology from 1918 to 1940, died after a sudden illness. The Department has thus not only lost its former leader, who was beloved by all its members, but suffered a serious loss as respects current research. Since his retirement Dr. Streeter had continued to work in the laboratory, compiling the important series of "Developmental horizons" which has been mentioned annually in these reports for

several years. His immense knowledge of human embryology, and his general wis-

dom, have been at the disposal of every worker in the laboratory.

PROGRAM OF INVESTIGATIONS

Morphological studies. Dr. C. H. Heuser, Curator of the Embryological Collection, devoted much of his time during the year to the preparation of about 20 valuable embryos in serial sections for addition to the collection. He also partly completed the study of a very interesting double monster, the earliest known specimen of this anomaly in man, which was contributed by Dr. John Gardner Murray, of Baltimore.

The Collection has received through Dr. A. T. Hertig three fetuses which were obtained as a result of deliberate termination of pregnancy because of strong indications from the maternal history that the fetuses would be abnormal. In one case the mother had German measles in early pregnancy; in another the pregnancy was preceded by birth of three successive Mongolian infants; and in the third there was a family history of Little's disease (congenital spastic paralysis). Dr. Heuser is now studying these interesting fetuses.

Dr. George L. Streeter completed during the year his study of the five oldest "horizons" or stages of development of the human embryo envisioned in the program described in the preceding Year Books. This brings his systematic account of the schedule of human development down to the end of the embryonic stage, after which the changes in external form and internal topography are less radical, and the size of the fetus is the only criterion necessary for estimation and comparison of age. In preparing his descriptions of the developing organ systems Dr. Streeter gave especial attention to the eye, ear, salivary glands, and kidney, and his comments on these organs represent

much new investigation as to the rate of their respective growth.

At the time of his death, Dr. Streeter had finished an article on horizons xix and xx, and had assembled all the illustrations for the next article, on horizons xxi, xxii, and xxiii. The notes on which he was to base the text on the three latter stages were left in rough draft. Dr. Chester H. Heuser has consented to see the former article through the press for the Contributions to Embryology, and to write the text for the latter on the basis of his knowledge of Dr. Streeter's intentions and his own thorough command of the subject matter. Embryologists all over the world who have been following the progress of Dr. Streeter's remarkable summary of human development will be happy to know that Dr. Heuser will continue the series, dealing with the earlier stages, with which he has had especially intimate experience.

Dr. E. Carl Sensenig during his summer visit with us studied the development of the enveloping membranes (meninges) of the human spinal cord, and of the joints and ligaments of the vertebral column. About 12 fetuses selected by Dr. Sensenig from the Collection, presenting various anomalies of the spinal region, have been serially sectioned for his use and will form the material for subsequent research.

Mention has been made above of the visit of Dr. C. M. Goss for the purpose of studying the embryonic human heart at the time of its earliest pulsation and the beginning of the circulation of the blood. Experimental observation by the methods used in rats being out of the

question for the human species, Dr. Goss could only examine prepared human embryos at stages of development spaced as closely as possible. His difficulties in this effort are of great interest with regard to the program of the Department. In brief, the Carnegie Collection of human embryos, incomparably the largest in the world, was not able to provide this experienced visitor with material sufficient for his quest. Dr. Goss, although he examined 61 embryos, could not obtain publishable evidence of the age at which the circulation of blood begins in man, simply because the specimens in the collection, of the crucial stage and preserved with the required perfection, are not sufficiently numerous. The lesson is that the addition of more and more embryos, of stages already fairly well represented, indicates not mere acquisitiveness nor the gratification of the collector's instinct, but in fact helps to make the collection more useful, because richer numerically, for the study of very critical and rapidly occurring events in embryonic life and for investigations that require comparison of many specimens and statistical analysis.

Mrs. Dorcas H. Padgett has continued work on the development of the cerebral veins in human embryos under a grant made to Dr. Corner by the Life Insurance Medical Research Fund.

Collection of very early human embryos and associated corpora lutea. Dr. Arthur T. Hertig, Research Associate, reports that his program of collection of human embryos continues in association with Dr. John Rock at the Free Hospital for Women, Brookline, Massachusetts, and the Boston Lying-in Hospital. Two phases of development are being sought especially, recent implantation and the stage of early yolk-sac formation. In order to find such stages Dr. Hertig has carefully studied the uteri removed at operation from 12

fertile, potentially pregnant patients during the past 12 months. Two early human ova were discovered, one about 12½ days old (Carnegie no. 8558), and the other about 17 days old.

Dr. Hertig is now studying intensively a collection of 27 human corpora lutea that were associated with early embryos found during the past 10 years. In this work he has the assistance of Mr. Roger White, a trained histologist who is at present a third-year medical student at Harvard. In addition to the study of these corpora lutea stained by the usual methods, histochemical studies are being made on the two early corpora lutea found this past year plus three more from somewhat more advanced pregnancies. Two of the latter were interrupted because of cancer of the cervix at 6 weeks and 4½ months respectively, and the remaining patient had heart disease which necessitated hysterotomy and sterilization at 10 weeks. During the coming year additional corpora lutea should be obtained in this manner. It is hoped that this histological and histochemical survey of the corpus luteum of early pregnancy will be ready for publication during the coming year.

Dr. John Rock, whose experiments on living human egg cells are done in association with Dr. Hertig's program, though not with direct Carnegie Institution support, reports that during the year 156 human ova were recovered from operative material and were cultured in vitro. Approximately 24 per cent of these ova showed one or two polar bodies after 24 hours of culture. One hundred and thirty of them were exposed to sperm suspensions. One ovum showed cleavage into two blastomeres, but was unfortunately lost. A similar specimen obtained about two years ago has been deposited in the Carnegie Collection.

Other primate species. Dr. Joseph Gill-

man, Research Associate, sent from Johannesburg during the year another early embryo of the baboon. This specimen, which is about 13 days old, is the youngest of that species yet obtained. Although there are signs of abnormality (old blood in the superficial chorion and recent free blood in the chorionic cavity), the specimen is apparently normal in general structure. It illustrates almost the earliest stage of yolk-sac formation. The Gillman series of very early embryos of *Papio porcarius* will form a valuable resource for the comparative embryology of the primates.

Experimental embryology. Dr. R. K. Burns spent practically all his available time working on two extensive reviews of the development of the reproductive system. Mrs. Arthur LaVelle (Faith Wilson), who has been at work under the direction of Dr. Burns, continued her studies of the development of the reproductive system of the hamster, an animal not unlike the rat anatomically, which has the shortest gestation period known in mammals (excepting of course the marsupials) and is therefore born with relatively undifferentiated sex organs. An abstract of part of her results, recently presented at a scientific meeting, will be cited below under "Published research."

Dr. S. R. M. Reynolds devoted the work of the year largely to the perfection of the tokodynamometer, which is described below in the summary of his publications, and in developing the application of the instrument to study of the human uterus during labor, in co-operation with the Department of Obstetrics of Johns Hopkins Hospital. This application of his physiological research, predicted in the previous Year Book, has proved immediately useful in the clinic even beyond expectation. To cite one example only, cases of delayed parturition which might have

been considered due to deficient uterine contractility are now explainable as the result of overactive contractions which lack the orderly co-ordination of rhythm and progression over the uterus that are necessary for effective emptying of that organ. Ways and means to relieve the abnormal condition may be planned on the basis of such an analysis of its exact nature.

Mrs. Lyla T. Bradin continued her investigation of the susceptibility of newborn animals to oxygen deprivation. It appears that the degree of susceptibility is an index of the degree of maturity of the young at birth. Mrs. Bradin has found it advisable to make comparisons between this index and other indices of maturity, for example the degree of ossification of the skeleton. She has therefore acquired the technique of "clearing" fetuses, that is to say, making the tissues semitransparent by dehydration and permeation with fluids of low refractive index so that the centers of ossification may be seen and studied under low magnification. A large series of cleared embryos of various species has thus been prepared, and many have been preserved permanently by the beautiful new method of inclusion in a block of transparent plastic.

Dr. Louis B. Flexner reports that the program of investigation of the morphological, biochemical, and physiological differentiation of the fetal cerebral cortex and liver has been continued with aid from the Committee on Growth, National Research Council, acting for the American Cancer Society, and from the U. S. Public Health Service. The phase-contrast microscope has been adapted with great effectiveness by V. B. Peters to a study of the histogenesis of the cerebral cortex. This instrument has permitted identification of the primitive axons and dendrites when they first sprout from the neuro-

blasts, and study of the subsequent development of their morphological pattern and changes in their refractive index. The method, in contrast with the capricious silver techniques formerly employed, has the great advantages of simplicity and dependability. The investigations of Josefa B. Flexner on a phosphatase believed to release energy for use by the cell from energy-rich phosphate compounds have shown that this enzyme begins to increase markedly in activity at the time when the neuroblasts first develop axons and dendrites. Her comprehensive survey of phosphorus metabolism reveals a remarkably constant quantity of energy-rich phosphate compounds per unit wet weight of tissue throughout development. The developmental pattern of respiratory enzymes, essential for the over-all metabolism of the cell, is being studied by E. L. Belknap, Jr. Experiments are also being made in an effort to understand the factors responsible for the characteristic absence of glycogen from the fetal liver, first noted by Claude Bernard in 1858. D. B. Tyler is assaying the pancreas for hormones possibly related to this phenomenon. W. Insull, Jr., and N. K. Proctor have shown that the enzyme phosphorylase, essential for glycogen formation, is highly active in all the developmental stages of the fetal liver, and that the absence of glycogen is therefore not due to absence of this enzyme.

Studies with the isotope-tracer technique on placental function and fetal nutrition in man have been carried out by L. B. Flexner, A. Gellhorn, and W. S. Wilde, of this Department, in collaboration with G. J. Vosburgh and L. M. Hellman, of the Department of Obstetrics, Johns Hopkins University and Hospital, and D. B. Cowie, of the Department of Terrestrial Magnetism. The subjects were patients admitted to the hospital for therapeutic

abortion or Caesarean section. The procedures involved are so simple and so completely innocuous that it can be confidently anticipated they will lend themselves to a variety of studies on placental function in normal and abnormal pregnancies. Those studies which have been completed with radioactive sodium and heavy water have demonstrated the close similarity in function of the human placenta with that of animals having the same morphologic type, have established a base line of normal placental function against which further observations on placental function in abnormal pregnancies can be evaluated, and have shown the huge protection afforded the fetus with respect to water and sodium (near term the human fetus receives across the placenta about 4000 times as much water and 1000 times as much sodium as is incorporated in growth). An article reporting part of this research is summarized below, under "Published research."

The comprehensive study of placental function and fetal nutrition in the guinea pig has been extended by Drs. Flexner, Vosburgh, and Cowie to include iron. Radioiron has been used to test the classic hypothesis that the fetus derives its iron from breakdown of maternal red blood cells in the placenta rather than from the minute amounts of iron in the plasma. Their results, which fail to support the classic view, were presented at the tri-annual meeting of the American Congress of Obstetrics and Gynecology, and are abstracted below under "Published research." The contribution was awarded the first prize offered by the Congress for excellence of the exhibit.

Experimental teratology; histology of the reproductive cycle. Dr. George W. Corner, Director, continued, as time permitted, his experiments on the production of high prenatal mortality in rabbits by

noxious influences applied to the maternal organism while the segmenting ovum is in progress through the Fallopian tube. The results, not yet ready for publication, have revealed a definitely harmful agency which may possibly be operative in certain human cases.

Dr. Joseph Gillman, Research Associate of the Department, with Miss Christine Gilbert, Mr. Theodore Gillman, and Miss Isobel Spence recently published a remarkable investigation on experimental teratology, done in their laboratories at the University of the Witwatersrand, Johannesburg, South Africa. These investigators find that injection of female rats shortly before mating and during early pregnancy with the well known colloidal vital dye trypan blue results in the production of a very high proportion of defective embryos showing congenital malformations such as hydrocephalus, spina bifida, eye defects, and deformities of the limbs.

Dr. Corner gave a good deal of time to the study of human ovaries sent by Dr. Edmond J. Farris, of the Wistar Institute of Anatomy and Biology, Philadelphia. These ovaries, or pieces of ovarian tissue, came from patients who were subjected to operation for gynecological diseases, and who had been studied by a test tentatively thought to indicate the occurrence of ovulation. This test, which was devised by Dr. Farris, depends on the presence in the urine of a substance which causes increased blood flow through the ovaries of test animals. The occurrence of ovulation can of course be confirmed, and an estimate formed as to the time of its occurrence, by examination of the ovaries of the tested patient if those organs happen to become available for microscopic study through removal at operation soon after the test. The detailed descriptive histology of the corpus luteum necessary

for estimation of its age has not been accurately worked out from human ovaries, but Dr. Corner has completed such studies in monkeys (see recent Year Books for references to the publications). Hence the human ovaries obtained by surgeons co-operating with Dr. Farris, in positive cases, and some negative cases as well, were submitted to Dr. Corner for comparison of the corpora lutea, maturing follicles, etc. with those of rhesus monkeys of known cyclic stage. The study has afforded a good deal of experience in judging the age of early human corpora lutea. It was found that the Farris test is positive not only when normal ovulation occurs, but also in connection with certain ovarian crises which lead to atresia of the advanced follicle rather than to ovulation. An illustrated description of the ovarian histology presented by these cases of late follicular atresia will be published later.

Co-operative activities. The co-operative associations with Dr. A. T. Hertig and Dr. John Rock, of Boston, with the Johns Hopkins ophthalmological clinic, and with the Department of Terrestrial Magnetism of the Carnegie Institution have been mentioned in the foregoing pages.

Co-operation with the Department of Obstetrics of Johns Hopkins University and Johns Hopkins Hospital, led from our side by Dr. S. R. M. Reynolds, has been especially active during the year. This also has been described at length in the foregoing section.

The monkey colony and the facilities for operative and other experimental work with monkeys have been used by special arrangement for work by Dr. Roger B. Scott, of the Department of Gynecology, Johns Hopkins Hospital, who is studying the experimental production of endometriosis, a disease of the female reproductive system which seems to be growing more common and which offers baffling

theoretical as well as practical problems in a field closely related to the Department's interests. Monkeys in use for experimental work have been housed for Dr. C. F. Geschickter, of Washington, under a long-standing arrangement, and temporarily for Dr. A. Earl Walker, professor of neurosurgery in Johns Hopkins University.

The Department receives many requests for the loan or donation of embryological material to workers in other institutions. These are granted when possible, especially when the investigator making the request has previously worked in the Department, or when he can come to the laboratory to start his work on the Carnegie material, so that there is mutual understanding of the project. During the

year 1947-1948 a number of fetal hands and feet have been made available to Professor Harold Cummins, of Tulane University. Two sets of sections of the fetal pelvis were lent to Dr. R. M. H. Power, of McGill University. Professor John F. Huber, of Temple University, visited the Department several times to study fetal lungs and has had the use of several series of sections at his own laboratory. Professor Ernst Van Campenhout, of Louvain, Belgium, visited the Department for several days to study primate gonads and was granted the loan of several sectioned ovaries of primates. Numerous other visitors came for brief visits or for a look at the unique collection of early human embryos.

THE CONTRIBUTIONS TO EMBRYOLOGY

Volume 32 of the Contributions to Embryology was published March 30, 1948. Copy for volume 33 was in press at the end of the year covered by this report (June 1948). The volume should be published within the calendar year 1948. The table of contents of volume 33 follows:

- 213. S. R. M. Reynolds, *Adaptation of uterine blood vessels and accommodation of the products of conception.*
- 214. E. Carl Sensenig, *The early development of the human vertebral column.*
- 215. Adolph H. Schultz, *The palatine ridges of primates.*
- 216. Somers H. Sturgis, *Rate and significance of atresia of the ovarian follicle of the rhesus monkey.*

- 217. Clara Eddy Hamilton, *Observations on the cervical mucosa of the rhesus monkey.*
- 218. Lawrence R. Wharton, Jr., *Double ureters and associated renal anomalies in early human embryos.*
- 219. Elizabeth Mapelsden Ramsey, *The vascular pattern of the endometrium of the pregnant rhesus monkey (Macaca mulatta).*
- 220. George L. Streeter, *Developmental horizons in human embryos (fourth issue): A review of the histogenesis of bone and cartilage.*
- 221. Arthur T. Hertig and John Rock, *Two human ova of the pre-villous stage, having a developmental age of about eight and nine days respectively.*

PUBLISHED RESEARCH

A bibliography of the published writings of the members, research associates, fellows, and guests of the Department will be found, as usual, at the end of this report. Some of the researches there listed have already been summarized in preceding

Year Books. Those not yet reported are summarized herewith.

A CASE OF QUADRUPLETS

Late in 1946 a good deal of popular interest was aroused through the report in

Baltimore newspapers that a case of quadruplet gestation had been recognized by X-ray. The obstetrician in charge of the case, Dr. Thomas S. Bowyer, readily agreed to a proposed co-operative study of the medical and biological aspects of the case. When delivery occurred, prematurely and precipitately, on December 23, at St. Agnes Hospital, Baltimore, the placentas and membranes were carefully tagged to correspond to the infants, and were given to Dr. George W. Corner for study. As it turned out, the case was of the variety which is least interesting from the scientific standpoint, for the quadruplet infants (three boys and one girl) each had a separate placenta and chorioamniotic membrane, although three of the placentas had become fused. The infants presumably, therefore, originated from four separate ova. A report by Dr. Bowyer and Dr. Corner, illustrated with the X-ray picture from which diagnosis of quadruplets was made at 200 days, and with excellent photographs of the placentas by Mr. Reather, was published in the *American Journal of Obstetrics and Gynecology* (see bibliography).

HOW LATE IN PREGNANCY IS THE CORPUS LUTEUM NECESSARY?

There is ample experimental evidence that the hormone of the corpus luteum, which originates at the site of an ovarian follicle after the ovum has been discharged, is necessary to the successful nutrition and uterine implantation of the fertilized ovum. Just how long the corpus luteum remains necessary to the welfare of the ovum is, however, a much discussed problem. In many of the small laboratory animals, which have relatively short gestation periods, it has proved impossible to remove the corpora lutea at any stage of pregnancy without causing death and

resorption of the embryos. There are, on the other hand, reports indicating that the human corpus luteum, and indeed the whole of both ovaries, may be removed quite early in pregnancy without disturbing the health of the fetus. About 1936 Dr. Carl G. Hartman, then a member of the Department of Embryology, began to investigate the problem in the rhesus monkey, and after his departure Dr. Corner continued the study as occasion offered. The final tabulation shows clearly that in this species the corpus luteum of pregnancy may be removed as early as the 25th day without disturbing the pregnancy. To carry the experiment farther, into earlier stages, would be very expensive because a reliable diagnosis of pregnancy cannot always be made before the 25th day, and also because it is to be expected that about the border-line period some operations would result in abortion and others not; a large enough number of animals to give statistical evidence would therefore be required.

One of the oöphorectomies done on the 25th day had curious and instructive results. The first ovary had been removed, presumably totally, a couple of years before this experiment; and it was only the second ovary that was excised by the present experimenters. The corpus luteum of pregnancy was removed in its entirety with the second ovary. The animal retained her fetus, delivered it on time, and then resumed menstrual cycles and maintained a high sex-skin color. In spite of this strong evidence that an ovarian fragment had been left behind, no such fragment could be found by careful search at three successive operations (under full anesthesia) done by Dr. Corner and Mr. Rever, both of them highly experienced in such work. Only when the uterus was removed and cut into bits was an ovarian fragment discovered, closely applied to

the uterus under dense adhesions, which had been left behind at the time of the first oöphorectomy. This experience illustrates the danger of assuming that surgical removal of the ovary has been complete, and has led Hartman and Corner to set forth, in their paper, strict criteria for experiments in which total oöphorectomy must be assured.

DEVELOPMENTAL STAGES OF HUMAN EMBRYOS

The third large installment of Dr. George L. Streeter's survey of the developmental stages or "horizons" of the human embryo was published in volume 32 of the *Contributions to Embryology*. It deals with horizons xv to xviii, covering the ages (dated from ovulation) from about 30 to 38 days. A summary of this article was printed in Year Book No. 44 (1944-1945).

DEVELOPMENT OF ACCESSORY REPRODUCTIVE ORGANS IN HAMSTERS

Mrs. Faith Wilson LaVelle, who is working under the direction of Dr. Burns, reported her results in the April 1948 abstracts of the American Association of Anatomists. A sudden acceleration of growth of the testes occurs at days 25 to 27 (postnatal), and of the male accessory glands and phallus at days 27 to 29. The female reproductive organs show similarly timed though slighter increases between 27 and 29 days, when the ovarian follicles markedly enlarge. Castration of day-old male hamsters results in repression of the accessories, whereas most of the female organs grow at approximately the normal rate after castration, until about 21 days, when growth levels off. These results, and also experiments on the response of immature structures to testosterone, demonstrate that the testis in-

fluences development of the accessories from birth on, whereas the ovary is not necessary until nearer puberty.

The normal postpubertal adrenal gland is larger in the male than in the female, but with castration the gland diminishes in size in the former and enlarges in the latter. Adrenocorticotrophin, in castrate and intact animals, maintains adrenal size at a level somewhat above normal in both sexes.

The golden hamster has dorsolateral skin spots which in males become conspicuous by growth of coarse black hairs in the spots. The skin in these spots begins to be pigmented at about 31 days in castrates and intact controls of both sexes. In normal males the local hairs soon begin to coarsen and blacken, but castrate males and all females retain soft, gray hairs over the pigment spots.

MIGRATION OF THE HUMAN GERM CELLS

One of the most striking general concepts of biology is that which considers the germ cells as having a lineage distinct from that of the other body cells or somatic cells. In some invertebrates, indeed, it has been possible to trace the descent of the germ cells from one of the first two cells produced when the fertilized egg divides, so that the egg cell gives rise to one set of cells of germ lineage, and to another set that clothe the germ cells, so to speak, with the animal body that is to carry them for one generation. In mammals no such complete separation of the two lineages has been traced as yet, but in some species the germ cells are at least set aside from the somatic cells in early embryonic life, even before the testes or the ovaries have developed. The germ cells first appear in primitive connective tissue along the axis of the body in or near the base of the mesentery, and mi-

grate into the gonads after the latter develop. The tissues of the gonads, according to this concept, do not give rise to the earliest germ cells, but act only as a place of reception and growth for them.

Such a story has been held true even of the human species. In 1911 and 1912 European investigators first described primordial germ cells outside the gonads in early human embryos. Subsequent accounts have been limited in value because of inadequate material. Professor Emil Witschi, of the University of Iowa, made this problem the object of a special investigation during a stay in the Department of Embryology in the summer of 1943. In the extensive collection of well preserved early embryos at his disposal Dr. Witschi was readily able to discern and follow cells which he identified as primordial germ cells. He studied in detail 23 embryos from the stage of 13 somites (group xi) to 8 mm. crown-rump length (group xvi). In the youngest of these embryos all the germ cells were in the endoderm of the yolk sac near the allantoic evagination. Dr. Witschi supposes that they arise either from the endoderm or from primitive cells which are the common source of both. In later stages the germ cells leave the endoderm and move into the mesenchyme. In embryos of more than 25 somites they leave the gut, ascend through the mesentery, and advance laterally toward the mesenteric folds. Dr. Witschi describes them as having pseudopodia and believes that they migrate by active ameboid movement. The germ cells reach their final position in the cortex of the primitive gonads by diverse routes and are believed to be oriented in their movement by some specific stimulus emanating from the gonadal peritoneum (prospective cortex of testis or ovary). After their arrival in the gonadal folds the germ cells become en-

closed in individual follicle cells (in the ovary) or in the testis cords, and their autonomous movement ceases. During the migratory period the number of germ cells increases by mitotic division.

Dr. Witschi found no facts and no convincing theoretical arguments in favor of secondary germ-cell formation, that is, the start of a new lineage of germ cells from somatic cells, as has been described in certain invertebrates and even in vertebrates. He is careful, however, to point out that further investigation will be necessary to settle finally whether the germ cells that he has traced as a separate line from an early stage to the definitive gonads are actually ancestral to all the sperm and egg cells of the adult.

DEVELOPMENT OF THE HUMAN TESTIS AND OVARY

The development of the gonads has offered a difficult problem. The general histological pattern of these organs is determined very early in development, and there are great subsequent changes in details of cell structure. For these reasons it is not a simple matter to establish with certainty the origin of any cell in the ovary or testis. The problem, however, underlies many of the most important questions in anatomy and pathology, including for example that of the lineage of the germ cells, mentioned in the preceding section of this report. The very complex problem of the classification of ovarian tumors will only be fully understood through knowledge of the embryological development of the ovary. The difficulties in the way of investigation, great enough in the case of laboratory animals, are even greater in man because of the scarcity of well preserved embryonic material. The Carnegie Collection is able because of its large size to supply the needed material in some

degree; and it is supplemented by a small but choice collection of embryonic human ovaries made by Dr. T. R. Forbes, now of Yale University, which has been left on deposit in the Department.

Advantage of all this material was taken by Dr. Joseph Gillman, of the University of the Witwatersrand, a visitor in 1942, whose monograph in volume 32 of the *Contributions to Embryology* is a comprehensive study of the development of the human testis and ovary. The article is so extensive and so full of descriptive detail as almost to baffle summarization in this report. All that can be undertaken here is a sketch of its contents.

Dr. Gillman points out first that the human gonad has a triple origin, from the coelomic epithelium, the underlying mesenchyme, and the primitive germ cells. The coelomic epithelium gives rise to the sex cords, the first buds of which form the rete complex. The mesenchymal primordium forms the supporting tissue and later receives an accession of cells from the general mesenchyme of the mesonephric body.

The testis begins to differentiate very early (14-16 mm. crown-rump length). The coelomic epithelium gives rise to the epithelium of the rete testis of the tubuli recti and of the seminiferous tubules (i.e., the Sertoli cells). The germ cells come from an extraneous source. The interstitial cells are derived from the mesenchyme. The rete cords, according to Gillman, are derived entirely from the coelomic epithelium; there is no evidence of derivation from the mesonephric tubules or from mesenchyme.

The human ovary develops from the indifferent gonad with less spectacular suddenness than does the testis. The young ovary consists of a covering layer of coelomic epithelium merging imperceptibly with the underlying cells derived

from the epithelium, which are called by Gillman pregranulosa cells. At about the stage of 140 mm. crown-rump length the primitive egg cells (oögonia) become encapsulated by pregranulosa cells, forming primitive follicles, and soon thereafter the mesenchymal stroma invades the ovary, dividing the medulla into strands or cords. There is a long period of interreaction between the invading stroma and the pregranulosa tissue, during which many oögonia degenerate, and highly variable patterns produced by incipient formation of follicles and degenerative reactions are seen. Ultimately the follicles become stabilized, with granulosa derived from the pregranulosa cells and thus ultimately from the coelomic epithelium, and with theca cells derived from the mesenchymal stroma.

Dr. Gillman's observations on the germ cells convinced him that they arise outside the gonad primordium. Thus the findings of Professor Witschi, mentioned above, agree with and notably complement those of Dr. Gillman.

The monograph must be read in detail in order to appreciate the author's views concerning such matters as the probable relation of the successive phases of growth and differentiation to endocrine influences in the developing embryo, the relation of gonad structure to sex inversion, and the histogenesis of tumors of the adult gonads.

DEVELOPMENT OF THE DIAPHRAGM

Dr. L. J. Wells, of the University of Minnesota, who spent part of 1947 in the Department's laboratory on a Guggenheim Fellowship, presented his preliminary observations on the development of the human diaphragm in the form of a demonstration at the 1948 meeting of the American Association of Anatomists. Dr. Wells reported that the development of

the diaphragm, which has been exceedingly difficult to observe and to describe, may be most easily understood (contrary to previous efforts at description) by first recognizing the mesenchymal primordium as a single entity with certain apertures. Thus, in embryos with closed pleuropericardial canals but open pleuroperitoneal canals, the apertures of the diaphragm are traversed by such obvious structures as the esophagus, aorta, and inferior vena cava, and also by the mesothelium of the right and left pleuroperitoneal canals. During subsequent closure of the latter canals, an invasion of the sites of closure by the mesenchyme of the primordium does occur. This invasion, however, is not an active phenomenon, but more a consequence of the growth of such subjacent organs as the liver and suprarenals. Thereafter the primordium differentiates into muscle. The definitive diaphragm, of course, is essentially a seromuscular septum between the thoracic and abdominal cavities.

EMBRYOLOGY OF A DIAPHRAGMATIC HERNIA

Congenital defects of the diaphragm are unfortunately not very rare. When they are accompanied by herniation of abdominal organs, for example liver, stomach, or intestine, into the pleural cavity, serious consequences ensue and elaborate surgical procedures are usually necessary. Opportunities for careful anatomical dissections are infrequent, and therefore every opportunity to study a congenital diaphragmatic hernia in any animal is useful, no matter how far the species is removed from the human. While Dr. L. J. Wells was in the Department in the spring of 1947, two officers of the Naval Medical Research Institute, Bethesda, Maryland (Commander R. B. Williams and Lieutenant J. H. Householder), asked advice about

a case encountered in a rat. Dr. Wells applied his expert knowledge of the embryology of the human diaphragm to the analysis of this case, deducing that it must have originated by abnormal headward growth of the right dorsal portion of the embryonic liver before closure of the right pleuroperitoneal canal. The case shows that an abnormal hiatus in the dorsolateral portion of the diaphragm is not necessarily due to nonclosure of the pleuroperitoneal canal.

DEVELOPMENT OF THE VERTEBRAE

Dr. E. Carl Sensenig, of the University of Alabama, who has been spending his summers recently in the Department, presented a report on his work at the April 1948 annual meeting of the American Association of Anatomists. He finds that in man, as in other mammals, the tissue which will give rise to the vertebrae forms at first a continuous medial concentration surrounding the notochord and lateral concentrations. This tissue has ventrolateral extensions which will contribute to the rib shafts, but otherwise the only indication of segmentation is given by the intersegmental arteries. Sclerotomic fissures then divide the medial concentration of mesenchyme into cranial and caudal sclerotome halves. Each of the primary vertebral centra develops from the caudal part of one sclerotome half and the cranial part of the next. The intersegmental vessels thus lie at the midpoints of the definitive vertebrae. A comprehensive and fully illustrated article on this subject by Dr. Sensenig is in press in the Contributions to Embryology.

DEVELOPMENT OF THE CRANIAL ARTERIES

A comprehensive article on the development of the cranial arteries, published in volume 32 of the Contributions to Embry-

ology, has an interesting history as well as valuable content. The author, Mrs. Dorcas Hager Padget, was trained as a medical illustrator and served in that capacity with the late Dr. Walter S. Dandy, the celebrated neurological surgeon of Johns Hopkins Hospital. Difficulties in operations on the brain, arising from unexpected variations of the cerebral blood supply, led Dr. Dandy to interest himself in the embryological development of the cranial arteries. It became Mrs. Padget's duty to make original studies on embryos in the Carnegie Collection, from which she made graphic reconstructions and ultimately definitive drawings of the embryonic arteries. Her large series of careful drawings, competently done from both the scientific and the artistic standpoint, is presented with a descriptive text outlining the complex history of the human cranial arteries. The results are not susceptible of summarization here. The development of the arteries of the head is characterized, like that of other parts of the blood vascular system, by changing patterns occasioned by the elimination of certain vessels which serve temporary needs, and the concomitant elaboration of new channels. The present study is concentrated on the formation of the various carotid and basilar branches, and includes the arteries supplying the internal ear and the eye as well as the brain. It will therefore be of interest to otologists and ophthalmologists as well as to neurosurgeons.

THE MAMMARY GLAND OF THE MONKEY

During the academic year 1939-1940 Dr. Harold Speert, then a recently graduated physician, was a guest in the Department of Embryology. He spent the year under the guidance of Dr. Carl G. Hartman making observations and experiments on the mammary gland of the

rhesus monkey. Dr. Hartman put at Dr. Speert's disposal a large collection of specimens acquired during his own long researches on the physiology of reproduction in the monkey. In subsequent years, while Dr. Speert was a hospital resident and afterward medical officer on military service, he continued the study of his preparations as time permitted. The result is an extensive monograph in the Contributions to Embryology on many phases of the embryology, anatomy, physiology, and pathology of the mammary gland, illustrated by 19 plates comprising 118 photographs made by Mr. Chester Reather. Among the significant facts as summarized by Dr. Speert, the following may be emphasized:

Pronounced individual variation was found in the mammary glands of both male and female monkeys under identical physiologic and experimental conditions. Changes in the gland were therefore studied by means of repeated biopsies in the same animal.

Cyclic changes occur in the mammary glands of mature females, in relation to the menstrual cycle. Lobular enlargement, resulting from dilatation of the alveoli, was observed during the premenstruum and gradually subsided during the postmenstrual period. This change occurs in association with ovulatory cycles; no mammary changes were observed when ovulation failed to occur.

Removal of the corpus luteum 5 days after ovulation resulted in a rapid shrinkage of the mammary lobules. Hysterectomy produced no effect on the mammary gland.

During the first 2 months of pregnancy a high degree of individual variation was found in the mammary gland, little growth occurring during this period. During the 3d month there is a sharp increase in the rate of glandular development.

Secretory activity is usually well established microscopically by the end of the 4th month of pregnancy. Mitotic proliferation of the gland cells continues to term. At term the mammary gland is uniformly developed; no "virginal lobules" remain.

Removal of both ovaries as early as the 31st day of pregnancy has no effect on the breast; glandular development and lactation proceed normally.

Secretory changes occur in the gland within 2 days after termination of pregnancy at 60 days.

Postpartum and postlactational involution of the mammary gland occurs irregularly, progressing at different rates in different lobules, in contrast with the uniform development of the gland during pregnancy. On the average, the lobules remain engorged for about 2 weeks after parturition or after the cessation of lactation. Involutionary processes become evident during the 2d week and are complete at the end of 3½ months. Involution appears to progress more rapidly in animals which are permitted to nurse their young than in those whose young are taken from them at birth.

After fetal death in utero, mammary involution begins irrespective of expulsion of the fetus. Involutionary processes were observed following the interruption of pregnancy as early as the 36th day.

Stimulation of mammary growth was produced by estrogens, progestins, androgens, gonadotrophins, and desoxycorticosterone acetate. Each hormone caused the same general pattern of response, which consisted of hyperplasia, dilatation, extension, and branching of the ducts, and growth of the lobules by multiplication and dilatation of the alveoli. Complete glandular development was produced and maintained in castrated females for periods up to 30 months by continuous

treatment with estrogenic injections and subcutaneous implantations of estrogen pellets. With 5 mg. of estradiol daily, complete glandular development was produced in prepubertal animals in 70 to 85 days. Stilbestrol, in increasing amounts up to 2 mg. per day for a month, produced no effect.

Mammary development was produced in castrated females by the sequential action of an estrogen (Amniotin) and progestin or anhydrohydroxy-progesterone. Crystalline progesterone, in daily doses from 5 to 20 mg., caused mammary growth in castrated females without estrogenic priming.

Testosterone and testosterone propionate, in amounts ranging from 5 to 50 mg. per day, each produced mammary growth in castrated and intact females. Mammary atrophy occurred when treatment was discontinued. Androstenediol and methyl testosterone (by mouth) were each without effect. Positive mammary responses occurred when testosterone or methyl testosterone was administered concurrently with an estrogen. When estrogen and androgen were administered sequentially, positive responses were obtained with Amniotin together with testosterone, testosterone propionate, or testosterone dipropionate, but not with methyl testosterone (by mouth). Epithelial proliferation which resulted in heaping of the cells and the formation of microscopic papillomas in the ducts and alveoli occurred in 2 females which received large doses of testosterone and testosterone propionate respectively.

Desoxycorticosterone acetate, in a daily dose of 10 mg., produced mammary development in castrated females.

Topical application of an alcoholic solution of estrone to one nipple area of prepubertal male monkeys resulted in localized, unilateral growth of the nipple and areola and the underlying mammary

tissue; whereas the control gland, treated with alcohol, was unaffected. These observations offer no support for the theory of pituitary mediation of estrogenic effects upon the mammary gland. Similar treatment caused no nipple growth in pregnant animals or in those injected with large amounts of estrogen.

PHOSPHATASE IN THE OVARY

Volume 32 of the Contributions to Embryology contains an article by Dr. George W. Corner on the presence and distribution of alkaline phosphatase in the ovarian follicle and the corpus luteum. The conclusions were summarized in the annual report for 1944-1945, Year Book No. 44. Publication of the article now makes available the photographic illustrations by Mr. Reather from the excellent microchemical preparations of Mr. Drane.

SPIRAL ARTERIES IN THE HUMAN OVARY

In last year's report (Year Book No. 46) reference was made to the description by Dr. Reynolds of a previously unnoticed fact of considerable theoretical importance concerning the course of the ovarian artery. In the rabbit this artery has a conspicuously spiral course as it runs through the hilum of the ovary. Following up this observation, Drs. Barnet Delson and Samuel Lubin, of the Cumberland Hospital, Brooklyn, New York, joined forces with Dr. Reynolds in studying the course of the human ovarian artery. Specimens from the operating room and from post mortems were injected through the artery with vinylite plastic and subsequently subjected to digestion. As in the earlier observations in the rabbit, the blood vessels were thus revealed, free of the rest of the ovarian structures, as beautiful branching trees of colored plastic. The course of the

ovarian artery through the hilum of the ovary was found to be distinctly spiral. The observation confirms and amplifies a brief description of the human ovarian artery given by an English investigator, Arthur Farre, 90 years ago. The collection and preparation of material in Brooklyn was aided by a grant from the Kate Lubin Research Foundation, Inc.

During the course of the study, in which about 60 pairs of human ovaries were injected, certain physiological deductions have been made. It is clear, for example, that the spiral arteries are highly developed, dense, and numerous only in the presence of estrogen. When the ovaries become sclerotic, the spiral arteries are few and poorly developed. Preparations from fetal and neonatal ovaries show well developed spiral arteries only at the time of birth at full term, when maternal estrogenic hormones are presumed to have been exerting a trophic action on the genital tract of the female infant. Part of this work has occasioned the award of the 1948 Foundation Prize of the American Association of Obstetricians, Gynecologists, and Abdominal Surgeons to Dr. Delson, one of the Brooklyn collaborators.

GROWTH OF THE PREGNANT UTERUS IN THE RAT

At the April 1948 meeting of the American Association of Anatomists, Mrs. Lyla T. Bradin presented her study of the relation between changes in weight of the products of conception and changes in shape and weight of the uterus during pregnancy, in the Norway rat. In these experiments, which were begun under the direction of Professor C. D. Turner at Northwestern University and continued with the advice of Dr. Reynolds, the animals were rendered sterile in one horn of the uterus by removal of the oviduct,

so that comparisons could be made between the pregnant and the nonpregnant horn of the same uterus. It was found that the most rapid fetal growth occurs during the latter third of the 21-day period of gestation. Before the 17th day the individual gestation sacs are cylindrical; after that date the uterus becomes cylindrical by elongation of the sacs. The pregnant uterus grows largely as a result of the mechanical stimulus of the growing products of conception. From the growth curves of the pregnant and the sterile horns respectively, it may be deduced that hormonal factors act to control the growth of the uterus adaptively so as to permit the products of conception to develop normally within the uterus. These results are in agreement with the analysis previously made of similar phenomena in the rabbit, by Dr. Reynolds.

STRUCTURE OF THE UTERUS IN RELATION TO MENSTRUATION

The phenomenon of menstruation occurs characteristically only in the human species, in the anthropoid apes, and in the Old World monkeys (macaques, baboons, etc.). Cyclic uterine bleeding of less conspicuous nature, being very slight or entirely internal, occurs in certain of the New World monkeys and in a few little-known animals considered to be related to the primates. It is therefore important to study every aspect of the uterus in these menstruating animals in comparison with those that do not menstruate. Previous observers have pointed out the striking fact that in the two best-known species in which menstruation is characteristic (man and rhesus monkey), the lining of the uterus (endometrium) is supplied with blood through small terminal arteries which are coiled into spiral form. It has been established that in the rhesus monkey

the coiled arteries undergo characteristic alterations during the menstrual cycle.

During his year (1946-1947) in the Department of Embryology, Dr. Irwin H. Kaiser prepared a description of the microscopic appearance of the coiled arteries in the rhesus monkey, and also studied three other menstruating primates (baboon, gibbon, and chimpanzee), in all of which he found coiled endometrial arteries. In the rhesus monkey the coiled arteries were found to pass through a regular series of alterations during the menstrual cycle. The occurrence of ovulation and consequent formation of a corpus luteum is followed by an increase in the number of coils, by special changes in the cell structure of the coiled vessels, and by shifting of the coiled parts of the arterioles toward the surface of the uterine lining.

Dr. Kaiser's article was awarded the Foundation Prize for 1947 of the American Association of Obstetricians, Gynecologists, and Abdominal Surgeons.

In a second paper Dr. Kaiser showed that in certain South American monkeys of the genera *Cebus*, *Ateles*, and *Alouatta*, which show cyclic changes in the lining of the uterus considered to be of menstrual nature, coiled arterioles do not exist. Dr. Kaiser suggests therefore that alterations of the coiled arterioles cannot be primarily responsible for the phenomenon of menstruation, although it is possible that the coiled arterioles regulate the amount of hemorrhage and tissue loss once the menstrual process is established.

PHYSIOLOGY OF ENDOMETRIAL GRAFTS

One of the most characteristic effects of the "female sex hormones" or estrogens is to cause dilatation of the blood vessels of the uterus. The hypothesis has won fairly general acceptance that this effect is caused by the local production, in the

uterus, of the vasodilating substance acetylcholine, under the action of the estrogens. Dr. Kaiser has reinvestigated the question, using rabbits in which a small piece of the endometrium had been grafted and made to grow in the anterior chamber of the eye, where it could be observed directly through the transparent cornea. In such animals estrogens produce a dilatation of endometrial vessels in the eye graft as well as in the uterus itself. The administration of atropine does not alter the response of the endometrial vessels to estrogens. Inasmuch as atropine is a powerful inhibitor of acetylcholine, the effect of estrogens upon the blood vessels of the endometrium must be exerted, as Dr. Kaiser states, through some chain of events not involving the action of that substance. These observations do not, however, exclude the possibility that the response to estrogens of vessels in the myometrium may be affected by atropine. It is known, moreover, that estrogens depress the cholinesterase in blood and tissue, thus producing in some circumstances a paradoxical cholinergic action.

PHYSIOLOGY OF THE UTERUS; TOKODYNAMOMETER

Dr. Reynolds, with the collaboration of Mr. O. O. Heard, of the Department of Embryology, and of Dr. Paul Bruns and Dr. L. M. Hellman, of the Johns Hopkins Department of Obstetrics, has produced a very great improvement in apparatus for recording the movements of the human uterus during labor. His instrument, which he calls a tokodynamometer, has the advantage of recording from three points on the pregnant uterus (upper part of fundus, middle, and lower or cervical region) and of responding to strictly local changes of form due to contractions of the uterine muscle. Previous instruments have re-

corded from one point only, and some of them, including the most commonly used, have caused errors due to changes of abdominal form produced by internal distention, etc. The new device consists essentially of three pick-up units and a direct three-channel recorder. The pick-up unit is a heavy metal ring with an opening 5 cm. in diameter. This ring is taped to the abdomen over the uterus. In the center of the ring a ball-pointed plunger also rests lightly on the abdominal skin. Any movement of the uterus below the skin changes the curve of the abdomen and causes a change in pressure on the plunger. The movement of the plunger with respect to the ring, which is less than 0.0015 inch, acts upon a small strain-gauge recorder fastened to the ring. The resultant change of electrical potential is recorded in a frictionless system on a moving strip of paper. Records from the three points, thus obtained without discomfort to the patient, are written as three tracings on the paper, from which the functional state of the muscular wall of the corresponding part of the uterus can be deduced. The records can be analyzed for frequency of contractions, rhythmicity of contractions, relative total work per unit of time, and characteristics of contractions such as intensity, duration of contraction and relaxation, rate, and gradients of activity. The spread of the contraction wave over the uterus can be followed as it reaches each of the three points.

The instrument has already proved itself useful in the obstetrical clinic, not only for research, but also for the practical understanding of cases of abnormal labor.

EFFECT OF ANTI-HISTAMINICS ON THE UTERUS

Dr. Reynolds and Dr. Kaiser have published a description of the method of

using the highly sensitive strain-gauge dynamometer for measuring the activity of the uterus in laboratory animals under anesthesia. The technical details will be found in the article. The method permits ready determination of the effect of drugs on the tone and rhythmic contractility of the uterus. The readings may be calculated according to formulas worked out by the authors, as percentage changes in activity (tone, contractility, total work) or as dynes per gram of uterine muscle per unit of time. The quantitative effects of Pitocin and of the anti-histaminic drugs Pyribenzamine and Benadryl are cited. The effect of the latter, when administered intravenously, is oxytocic, tone and rhythmic contractility being equally affected. The distended pregnant uterus is affected in the same way as the nondistended portion of the same uterus. The hormonal status (pregnancy, nonpregnancy) of the rabbit does not affect the response of the uterus to Pyribenzamine.

THE SOURCE OF FETAL IRON

Dr. Flexner reported at the 1948 meeting of the American Association of Anatomists the results of a study, made with the collaboration of Dr. Vosburgh and Dr. Cowie, of the sources of the iron received by the developing fetus. The generally accepted view has been that the fetus derives its iron from disintegrated maternal red blood cells. The contrary view that the iron which reaches the fetus comes from the plasma of the mother's blood rather than from her red blood cells has been difficult to accept because the amount of iron available in the plasma is very small (about 1 microgram of iron per milliliter of plasma). In the present experiments radioactive iron was administered to guinea pigs, which were thus caused to build the iron into ferric beta₁

globulinate in their own plasma. The guinea pigs were bled and the tagged plasma was injected into pregnant guinea pigs. The rate of supply of iron to the fetus was calculated from the radioiron found in the fetuses and placentas. The iron which reaches the fetus from the plasma, when compared with the amount of iron known to be required by the fetus in its growth, is found to be sufficient to meet the need. The opposite experiment, of administering red blood cells containing radioiron, is inconclusive because when the blood is given by transfusion, the red blood cells tend to hemolyze and thus lose the tagged iron to the plasma.

PERMEABILITY OF THE HUMAN PLACENTA TO SODIUM

An important contribution to our knowledge of the permeability of the human placenta has been published by a group of workers including Drs. Flexner and Wilde of the Department of Embryology, Dr. Cowie of the Department of Terrestrial Magnetism, and Drs. Hellman and Vosburgh of the Johns Hopkins Department of Obstetrics. This research was begun before the war by Drs. Flexner, Hellman, and Gellhorn. Tentative conclusions based on the first 10 cases were summarized in Year Book No. 43 (1943-1944). Since the war new cases have been studied to make up a total of 27. Choosing women in whom pregnancy was to be terminated for clinical reasons by abdominal hysterotomy, by Caesarean section, or by vaginal delivery, the investigators administered intravenously a small amount of sodium chloride made from sodium which had been rendered radioactive by bombardment in the cyclotron of the Department of Terrestrial Magnetism. This injection was done about 30 minutes before delivery of the fetus. The amount

of the tagged sodium that had reached the fetus, passing through the placenta from the maternal to the fetal blood stream, could then be measured directly in the fetal blood and tissues, and various deductions could be made from the amount transferred and the rate of transfer at the successive stages of pregnancy from the 9th week until full term. The technical procedures and methods of mathematical analysis worked out in animals have been described in several previous articles, as reported in recent Year Books.

Summarizing the results in the 27 human cases, the permeability of the placenta to sodium increases about 70 times from the 9th week to term. The permeability to sodium in the human placenta is of the same order as that in other animals having similar (hemochorial) placentas, and is much higher than in animals having placentas of the other types (epitheliochorial, syndesmochorial, and endotheliochorial). Once again, as the authors remark, Grosser's classification of placental types on the basis of microscopic structure proves to be significant physiologically as well as morphologically.

In a small series of cases, chronic vascular hypertension and cardiac disease were without effect on the rate of transfer. One of two cases of pre-eclampsia studied showed a marked reduction in permeability to sodium.

The relative amount of sodium reaching the human fetus across the placenta is very high in comparison with that in other animals. Expressed as the ratio of the amount supplied to a unit weight of the fetus to the amount retained by the unit weight of fetus in its growth ("safety factor"), the ratio in man is 160 at 12 weeks, and over 1100 at term. The safety factor in the sow, the lowest yet observed, is 3.5, and that in the guinea pig, the highest previously observed, averages 50.

PHYSIOLOGY OF THE EYE

Readers of this report will readily understand that research methods are not necessarily limited in their use to any one of the subdivisions of a scientific field such as biology. We have before us a good example of interchange between specialties, in the co-operative affiliation of Dr. R. O. Scholz, of the Wilmer Ophthalmological Clinic of Johns Hopkins University and Hospital, with Dr. Flexner and his associates. Through the mutual efforts of Dr. Scholz, Dr. Cowie (of the Department of Terrestrial Magnetism), and Dr. Wilde, the experience with the use of radioactive isotopes for the study of movements and exchanges of fluid within the animal body has been applied to the physiology of the eye, and in particular of the aqueous humor, the fluid which fills the anterior chamber in front of the lens. When it is recalled that abnormally high pressure of the aqueous humor is a major feature of the disease known as glaucoma, the importance of such an investigation is obvious.

Experimental studies by the group named above were begun several years ago. Two papers have now been published (see bibliography under Scholz and Wilde) in which certain basic principles and methods that have been worked out for use in the research are set forth.

The first of these articles deals with the ratio between the sodium present in the aqueous humor and that in the plasma of the circulating blood, from which the aqueous humor must of course derive its water and other ingredients. Sodium is one of the more important of these ingredients, and radioactive sodium is therefore one of the most important of the isotopes which must be used in studies of transfer rates. Measurement of the relative concentration of sodium in the

aqueous humor, in comparison with that in the plasma, which prevails when the system is in a "steady state" is obviously fundamental to subsequent experimental work in which altered rates of transfer are to be studied.

The observations were made by administering radioactive sodium chloride to guinea pigs, and after 24 hours withdrawing (under anesthesia) samples of the blood and of the aqueous humor. The average equilibrium ratio in the guinea pig was found to be 0.920 with a very small standard error.

It next becomes important to know how this ratio in the eye compares with that which exists between the blood plasma and other body fluids. Unfortunately no such value is available for guinea pigs. A ratio for dogs, worked out by other investigators, was found to be higher, namely, 1.12 when corrected for use in the present comparison. If this value is tentatively accepted as applying to the guinea pig, the lower ratio for the eye must be explained. Several factors may be operative, such as (1) a species difference, (2) concentration of sodium in the aqueous humor by loss of water into the tears, (3) secretion of sodium into the aqueous humor by the ciliary body or iris. It will be seen that by working out a precise method of investigation by radioactive sodium, the authors have raised a number of questions which must next be answered.

In their second article, the same authors begin an attack on the question whether active secretion of sodium occurs at the barrier between blood and aqueous humor, that is, in the ciliary body and iris. This problem may best be investigated by studying the rate of transfer of normally occurring constituents, and a good beginning can be made by following the rate of transfer of radioactive sodium.

When a readily diffusible substance such as ionized sodium is present in differing concentration on both sides of a barrier membrane such as the walls of the blood capillaries, it passes both ways. In experimental use of the tracer element, it is usual to build up a high concentration on one side (in this case, the blood, by intravenous injection), and to measure the tracer accumulated on the other side (in this case, the aqueous humor) as quickly as possible, in order to minimize the error introduced by countertransfer toward the blood. This method, however, involves certain errors and difficulties of its own. The authors have therefore worked out from logical mathematical considerations a double exponential equation which takes account of the outflow as well as of the changing inflow after intravenous injection. By use of such an equation, samples of the two fluids may be compared at convenient intervals after injection of the tagged substance, when the values in the fluid on the secondary side of the barrier are of greater magnitude and hence more accurately measurable than the smaller early concentrations that must be measured when the direct method is used.

DIFFUSION AND POPULARIZATION OF RESULTS

Dr. Corner gave a lecture in Minneapolis in October 1947, under the auspices of the Minnesota Human Genetics League and the Dight Institute for the Promotion of Human Genetics, on the embryology of twins and other multiple births. He also spoke before the University of Minnesota School of Medicine on the nature and causes of prenatal defects.

Dr. S. R. M. Reynolds took part, by invitation, in a panel discussion on gynecologic bleeding before the Section of Ob-

stetrics and Gynecology of the American Medical Association in June 1947. His contribution, which appeared in the *Journal of the American Medical Association* in November 1947, is a general survey of theories of the physiological basis of menstruation.

In February 1948 Dr. Reynolds took part in the annual symposium of the National Committee on Maternal Health, speaking on patterns of uterine contraction. In April he spoke at the Cumberland Hospital, Brooklyn, New York, on patterns of uterine contraction in pregnant women. In May 1948 he addressed several clinical societies and medical classes in Denver, Colorado, on problems of prematurity, on the peripheral circulation in the ovary, and on human tokography.

Dr. Reynolds also prepared for the *Annual Review of Physiology* a general review of the literature on the physiology of reproduction for the period May 1, 1946 to March 31, 1947.

Dr. Flexner, Dr. Vosburgh, and Dr. Cowie of the Department of Terrestrial Magnetism attended the annual Symposium on Quantitative Aspects of Biology of the Long Island Biological Association,

held at Cold Spring Harbor in June 1948. Dr. Flexner presented their paper on the use of radioactive tracers in studies of capillary permeability.

Dr. R. K. Burns prepared an extensive review on hormones and the differentiation of sex for a forthcoming book, "Survey of biological progress." He also wrote during the year a section on the urogenital system for a new comprehensive handbook of embryology to be entitled "The analysis of development."

Dr. Burns, Dr. Reynolds, Dr. Flexner, and Dr. Corner all gave one or more lectures each, by invitation, to classes in Johns Hopkins Medical School.

Mr. Chester Reather, departmental photographer, exhibited eight of his photographs at the International Exhibition of Biological Photography held at Rochester, New York, in September 1947. Four of these were selected for the traveling salon of the Biological Photographic Association.

The work of the Department was discussed in a radio broadcast at Padua, Italy, in March 1947 by Professor L. Muratori, who spoke on "L'Istituto Carnegie di Baltimore e i recenti progressi dell' embriologia umana."

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DEPARTMENT OF GENETICS

Cold Spring Harbor, Long Island, New York

M. DEMEREC, *Director*

Research at our Department is following the pattern outlined in our long-term program about three years ago, which concentrates efforts on the elucidation of fundamental problems about the nature of genic substances. This program was conceived on broad lines, because we believe that only by approaching the problem from several angles can we accumulate well balanced information on which to base conclusions. There is a definite advantage in having these diversified approaches represented at our laboratory, since only thus can our group derive early benefit from various discoveries and also avoid the pitfalls of narrow specialization.

The work of our group is yielding significant results. Discoveries made by McClintock suggest that what we have considered to be one gene may in some cases be composed of a number of re-duplicate units and may undergo relatively frequent changes in the number of its components, some of these changes showing up as well defined mutations. Our research with *Drosophila* and bacteria, using very sensitive methods to detect changes in genes, has suggested that genetic changes may be induced by many chemicals; and consequently, our concept of the gene as an entity resistant to external influences has had to be modified. Effects induced on chromosomes in the experiments by Kaufmann using combined treatments involving X-rays, ultraviolet, infrared, and nitrogen mustard have revealed that the final outcome of treatment is determined by several factors, among which biological factors play an important role. The histochemical re-

searches of Kaufmann and McDonald have provided a method for the localization of tryptophane-containing proteins within the chromosomes; and Caspari has been able to identify genes responsible for differences in the tryptophane content. Doermann's experiments on multiplication of bacteriophage are closely related to our central problem. Evidence obtained by several workers suggests that a certain degree of disintegration precedes multiplication of phages. The method developed by Doermann for studying phage growth may throw more light on the actual processes involved, and particularly on the mode of action of hereditary mechanisms in these processes.

As expected, collateral problems have developed during our studies. Investigations of one such problem are reported by MacDowell, who has found a very interesting relation between a virus infection and the behavior of certain lines of leukemia in mice. Another problem of this kind has been worked out by Witkin, in analyzing a case of population dynamics in cultures of two strains of bacteria grown together. This particular study is closely related to the wide range of problems investigated by Dobzhansky, which deal with the dynamics of natural populations and the factors responsible for organic evolution.

Investigations of the newly arising mutable loci in maize have been continued by McClintock. These loci fall into two classes, those requiring the presence of a second locus if instability is to be expressed and maintained, and those independent of such a locus. The

same controlling locus, designated *Ac* to symbolize its activator nature, is associated with mutability of all unstable loci of the first class. Not only have many previously unknown loci become unstable in the cultures studied, but some well known loci also have become unstable. There are independent occurrences of instability of the same locus. During the year, attention has been directed mainly to five *Ac*-controlled mutable loci. With respect to control of time and frequency of mutations, all five respond alike to *Ac*. Instability of an *Ac*-controlled locus is maintained only when *Ac* is present in the nucleus. When *Ac* is absent, the locus is completely stable. The most striking of the three main factors controlling the time and frequency of mutations of these loci is the dosage of *Ac*. The endosperm tissues are 3*n*. In these tissues, the effects of one, two, and three doses of *Ac* may be compared. Larger dosage delays the timing of mutations of all *Ac*-controlled mutable loci; the more *Ac* loci are present, the later the mutations occur. The *Ac* locus itself is mutable, the mutation being recognized by changes in the time and frequency of mutations occurring within the *Ac*-controlled loci. The mutations of *Ac* suggest that this locus is composed of a number of reduplicate units, and that a mutation at this locus involves an increase or a decrease in this number. Mutations of the *Ac*-controlled loci appear to be similar; they also suggest that the loci involved are composed of a number of reduplicate units, and that a visible mutation is the expression of an increase in this number. The mechanism responsible for such changes in units within a locus may be a form of unequal exchange between sister chromatids occurring in somatic nuclei. Experiments are under way to test the validity of this interpretation.

Histochemical studies have been con-

tinued by Kaufmann and his associates, using purified crystalline enzymes in combination with various staining procedures. Such methods have usually involved determination of the amount of stainable material removed during the period of enzymatic hydrolysis, but Kaufmann has now shown that a marked increase in color may result when action of the enzyme liberates a stainable component from a nonstaining compound. By application of this procedure a histochemical method has been developed for the localization of tryptophane-containing proteins. These studies have provided information concerning the *in situ* localization of nucleic acids and proteins as they exist independently and in combination, and the affinity of these materials for certain dyes.

McDonald's chemical studies have shown that the ability of ribonuclease to hydrolyze ribonucleic acid is greatly inhibited if desoxyribonucleic acid is present in the digestion mixture. The inhibition appears to be due to competition by the two nucleic acids for the enzyme, since the percentage of inhibition is greater the lower the concentration of the substrate (ribonucleic acid)—the concentration of the inhibitor (desoxyribonucleic acid) being constant—and is the same for any one ratio of the two acids. Salts affect the inhibition markedly.

Analysis of the modifying action of near infrared radiation on X-ray-induced gene and chromosome alterations in *Drosophila* has been continued by Kaufmann. The data indicate that the induced breaks are not all of one type, but fall into qualitatively different classes. Kaufmann has also extended these studies to determine the effect of such supplementary treatment on rearrangements induced by the chemical mutagen nitrogen mustard. It is shown that pretreatment with near infrared

radiation increases the frequency of nitrogen-mustard-induced rearrangements, but that posttreatment does not. This finding parallels the results obtained with X-rays. One striking difference in the action of these two mutagenic agents was revealed, however, by the finding that the frequency of rearrangements induced by nitrogen mustard increases with passage of time during the entire period that the treated males remain fertile, whereas there is a marked decline in frequency about 14 days after X-ray treatment. Kaufmann points out the great potentialities of the near infrared portion of the spectrum for an experimental approach to fundamental problems of chromosome organization, using intense monochromatic radiation of known physical properties.

Caspari has studied the action of the genes *a* in the meal moth *Ephestia* and *T* and *Fu* in the mouse. The gene *a* inhibits the production of kynurenin, a substance necessary for the formation of eye pigment, produced in the mammalian organism (mouse liver) from tryptophane. He has shown that *aa* proteins contain more tryptophane than *a⁺a⁺* proteins, and that their rate of autolysis is slower than that of *a⁺a⁺* proteins. The two facts agree in demonstrating qualitative differences in the protein make-up of the two strains. In extensive experiments it was impossible to demonstrate the presence of an enzyme converting tryptophane into kynurenin in *a⁺a⁺* and *aa* *Ephestia* homogenates and extracts. Furthermore, *a⁺a⁺* *Ephestia* larval testes do not transform tryptophane into kynurenin in vitro, although they produce kynurenin if transplanted to *aa* larvae. In the mouse, an antigen correlated with the gene *T* or with a gene closely linked to it has been demonstrated in sperm cells.

Investigations dealing with the mutagenic potency of carcinogens and chemi-

cally related compounds, carried on by Demerec and Wallace, have shown that chromosome breaks may be induced in *Drosophila* by four of the sixteen chemicals tested. Additional evidence has been obtained in support of the previously reported observation of a close correlation between mutagenicity and carcinogenicity. Studies of chemical mutagenicity conducted by Witkin with bacteria have added caffeine and colchicine to the list of three positive chemicals reported last year (sodium desoxycholate, acriflavine, and pyronin). Suggestive, but not yet conclusive, results have been obtained with several other compounds, including sodium chloride. Of the toxic compounds tested, only two—methyl green and basic fuchsin—were found to be nonmutagenic. All these results indicate that mutagenicity is a widespread property in chemicals.

Witkin has completed an analysis of the changes in population constitution that take place when two strains of bacteria (B and B/r of *Escherichia coli*) are grown together. She has found that during the period of rapid growth of the culture no appreciable change from the initial proportions occurs, but that as the culture grows older there is a rapid replacement of B/r by B. This is due partly to the greater ability of B bacteria to divide in old cultures, and partly to the more rapid death of B/r bacteria.

Doermann has devised a method for determining the number of bacteriophage particles present in an infected bacterium at any one stage after infection. Two main types of procedure have been developed: one in which the bacterial cell is lysed by an excess of a second phage in the presence of a metabolic poison, and another in which sonic vibration is used to disrupt the infected bacterium. The sonic experiments were carried out in collaboration with Dr. T. F. Anderson, of

the Johnson Foundation, University of Pennsylvania School of Medicine. With the lysis technique, it was found that two poisons, 5-methyltryptophane and cyanide, gave quantitatively different results. The sonic experiments agreed with those in which cyanide was used; and the conclusion was drawn that both the sonic-vibration procedure and the cyanide-lysis procedure give reliable estimates of the number of phage particles within the infected bacterium at any given time.

MacDowell reports that a virus, which now appears to have been transmitted for many years from mouse to mouse along with the cells of line-I leukemia, has been eliminated from a branch of this line by transmission through hosts previously immunized to the virus. Since these leukemic cells are transplanted every four days, and the sickness caused by the virus does not appear until after a week, this sickness was observed only when leukemic death was greatly delayed by the smallness of the dose, or prevented by natural or induced host resistance. The absence of this sickness when the decontaminated leukemic cells are used under the above conditions indicates that the virus was responsible for the sickness. Moreover, the successful resistance to virus-free leukemic cells induced by treatment with normal tissue from a certain strain (StoLi) and by treatment with actively immunized tissue from the host strain proves that the presence of this virus in the past was not responsible for the observed resistance.

Other lines of transplanted leukemia have yielded viruses that produce similar, but not identical, conditions in hosts of the same strain, and give cross-resistance with the virus from line I. By the use of virus-immunized hosts, three of these lines also have been decontaminated. The three cases of spontaneous leukemia tested were

virus-free. The facts that all mice of strain C58 have been susceptible when first treated with a virus, and that decontaminated leukemic lines can be maintained free of virus, indicate that the breeding colony is virus-free.

Professor Th. Dobzhansky, of Columbia University, Research Associate of the Carnegie Institution, has continued his studies of the genetic structure of natural populations, using chiefly two species of *Drosophila*. Again, he spent the summer working at the gardens of the Division of Plant Biology of the Institution, at Mather, California. He obtained additional evidence of seasonal changes in the composition of natural populations. Studies made under laboratory conditions have shown that the fitness for survival of various chromosomal types in populations is greatly influenced by temperature, which in this case serves as a mechanism of natural selection. He has also found considerable differences in fitness between various combinations of types. This means that a species having several types is readily adaptable to changing environments and can exploit the accessible environments more fully than can a homogeneous species.

We have been able to expand the research at our Department through grants obtained for work on specific problems. The U. S. Public Health Service has made funds available for support of Kaufmann's research dealing with the nature of changes induced in the living cell by irradiation; and the American Cancer Society, on recommendation of the Committee on Growth of the National Research Council, is supporting the work of Demerec dealing with tests of the mutagenic potency of various chemicals, particularly carcinogens. It has thus been possible to conduct experiments requiring laborious techniques

which otherwise would be difficult to undertake.

With the aid of funds available through the last-mentioned grant, Dr. Bruce Wallace has been added to our research group and Dr. Evelyn M. Witkin retained as a member. Until recently, Dr. Witkin held a fellowship of the American Cancer Society. Our Carnegie Institution fellows are a considerable asset to the group. Dr. A. H. Doermann has been with us all the past year, working on reproduction of bacteriophages. Dr. Berthe Delaporte arrived in June 1948 from the Centre National de la Recherche Scientifique in Paris, to study mutations and cytology in bacteria. In the fall of 1948 our group was joined by Dr. H. Clark Dalton, who will work on problems dealing with the genic control of pigments in animals. We await the arrival of Dr. G. Bertani, of the University of Milan, Italy, who will work on chemically induced mutations.

Through our collaboration with the Biological Laboratory two additional research members are associated with our group, Dr. V. Bryson and Dr. A. Kelner. Both are working with mutations in bacteria and other microorganisms. The Cold Spring Harbor Symposium on Quantitative Biology this year brought more than a hundred and fifty scientists to the Laboratory for a nine-day conference on the subject "Biological applications of tracer elements." A good opportunity was thus presented for our group to acquire first-hand information in this rapidly developing field of research. The conference covered a wide variety of topics. It included two papers dealing with results of experiments in which radioactive isotopes were used for study of cytogenetical problems. Six scientists came from Europe to

take part in the symposium program: G. Ehrensward, E. Hammarsten, and G. Hevesy from Sweden; H. H. Ussing from Denmark; R. Bentley from England; and K. Bernhard from Switzerland. During the summer, about fifty scientists and their assistants stayed at the Biological Laboratory.

Our Department has charge of a *Drosophila* Stock Center, where about 600 different stocks of mutants and combinations of mutants are maintained. These stocks are available to any scientist for research purposes. In 1940 a selected set of stocks, suitable for simple experimentation in genetics, was placed at the disposal of teachers in high schools and colleges. At the same time an illustrated pamphlet, the *Drosophila* guide, written by Demerec and Kaufmann, was published by the Institution. It describes the life cycle of *Drosophila melanogaster* and methods used in breeding and research with this fly, and gives an outline of simple experiments that can easily be completed by students. Since its publication it has passed through four editions and one reprinting. Mrs. Guinevere C. Smith is in charge of the *Drosophila* stock room. Last year about 300 cultures were sent to research workers of this country and of eleven foreign countries in Europe, Asia, Africa, and South America. Over 700 cultures were sent to teaching laboratories, mostly in high schools. That high-school students are taking advantage of this opportunity to experiment in genetics is further indicated by the fact that two recent winners of the Westinghouse Science Talent Scholarships used *Drosophila* in their research problems submitted to the awarding committee.

ORGANIZATION OF THE CHROMOSOME

B. P. KAUFMANN, M. R. McDONALD, H. GAY, K. WILSON, R. WYMAN, AND N. OKUDA

Our experimental approach to problems of gene structure and chromosome organization, originally formulated at the cytogenetic level, has since been supplemented by chemical and histochemical methods of analysis. The latter procedures have been stressed during the past year, as we have sought an understanding of patterns of organization at the submicroscopic level. Information concerning the localization of cellular materials and their distribution in the cycle of mitosis should aid in determining the nature of the changes induced in the living cell by ionizing and other types of radiation and by chemical mutagens. In these studies we are utilizing a variety of types of cells from a series of plants and animals. Progress of the work has been facilitated by a grant from the U. S. Public Health Service, which has provided us with additional scientific assistance.

HISTOCHEMICAL STUDIES

Preparation and assay of crystalline enzymes. Experiments on the preparation and assay of trypsin, chymotrypsin, pepsin, ribonuclease, and desoxyribonuclease for use in our histochemical studies of chromosome structure are continuing. The method described last year for the preparation of "protease-free" crystalline ribonuclease has been perfected and the results published. Dr. Kunitz, of the Rockefeller Institute for Medical Research, has recently succeeded in developing a long-awaited procedure for the preparation of crystalline desoxyribonuclease. Using his method, we are gradually accumulating a stock of this much-needed enzyme, for assay of its purity and enzymatic specificity and for subsequent use as

another specific histochemical tool. Unfortunately, the procedure is rather laborious and time-consuming, the yields are low, and the amounts of source material (pancreas) available to us have been extremely small and variable.

Localization of cellular materials. Enzymatic degradation has been used extensively in recent years for diagnostic purposes in cytological studies. Although crystalline enzymes have sometimes been used, they often contained traces of other enzymes whose presence precluded the formulation of unequivocal interpretations concerning the specificity of action of the major component. Enzymes also have frequently been used without assay of their own specific activity and without control of the numerous variables that can modify their action (e.g., time, pH, and temperature of digestion; concentration and stability of the enzyme; nature of the solvent; and condition of the substrate as determined by fixation and subsequent treatment). In this study only enzymes that, when assayed, have given negative tests for all possible interfering impurities have been used. Chemical assays of the activity of the enzyme at the beginning, during, and at the termination of all periods of hydrolysis have been made. A series of controls adequate to determine that variations in staining pattern are attributable to the enzyme and not to extraneous factors has also been maintained (Year Book No. 46 [1946-1947]).

This report concerns primarily the use of purified crystalline ribonuclease and pepsin, alone and in combination with each other, in association with various cytological procedures for the localization of ribonucleoprotein, ribonucleic acid, and nonhistone protein within the fixed cell.

Analyses, using combined enzymatic and staining procedures, have usually involved determination of the amount of stainable material removed during the period of enzymatic hydrolysis. Experiments in this laboratory during the past year have demonstrated that such processes may also result in a marked increase in color. The methods developed here have provided information concerning the *in situ* localization of nucleic acids and proteins as they exist independently and in combination, and the affinity of these materials for certain dyes.

Data obtained from studies of sections and smears of root tips of onion and lily and of salivary-gland cells of *Drosophila* and *Chironomus* serve to illustrate the results obtained.

1. Removal by ribonuclease of stainable material from chromosomes, nucleoli, and cytoplasm: If sections of root tips, fixed in Carnoy's acetic-alcohol, are stained in Unna's pyronin-methyl green combination, a pattern of differentiation is obtained in meristematic cells in which cytoplasm and nucleoli are red, chromosomes blue. In simultaneously stained sections that have been previously hydrolyzed in "protease-free" crystalline ribonuclease, the chromosomes are bright green and the cytoplasm and nucleoli are colorless. A similar pattern of differentiation and modification with ribonuclease is found in salivary-gland cells of *Drosophila* and *Chironomus*. We have attributed these results to the specific action of ribonuclease in degrading ribonucleic acid so that it is no longer stainable with pyronin.

Comparable results are obtainable with materials fixed in Helly's or Zenker's fluid. With fixatives of recognized superiority for cytological studies, such as Navashin's, or especially Flemming's (or its various modifications embodying chromic, osmic, and acetic acids), it is

difficult to secure good differentiation with pyronin-methyl green. Other dye combinations must be used. One of the most satisfactory of these is the safranin-gentian violet-orange G sequence (sometimes known as Flemming's triple stain), with which it is possible to obtain in mitotic plant cells a pattern of differentiation in which metaphase and anaphase chromosomes are bright red, the chromonemata of late telophases and early prophase are violet, nucleoli are red, and the cytoplasm is almost colorless, having a slight reddish or red-orange cast. Under identical conditions of differentiation, after ribonuclease treatment, there is a reduction in intensity of coloration with safranin, so that the condensed chromosomes stain violet instead of red, and the nucleoli less brilliantly than before digestion. Prolonged treatment with ribonuclease may also impair stainability with gentian violet so that the chromosomes become progressively paler. Concomitantly with this reduction of stainability with the basic dyes, enhanced coloration with orange G is obtained in chromosomes, nucleoli, and cytoplasm. These observations indicate that in addition to a constant ribonucleic acid component of the chromonematic threads, there is a more labile chromosomal ribonucleic acid that undergoes in the course of mitosis a cycle of accretion and dispersion coincident with the disappearance and reappearance of the nucleolus. This suggests that the nucleolus may serve as a reservoir of ribonucleic acid, which is gradually transferred to the chromosomes during the prophase. Accumulation of this material by the condensed chromosome may serve to provide a state of temporary heterochromatization, for we have found that ribonucleoprotein is a constant component of permanent heterochromatin. Because heterochromatic regions, such as the proximal third of the

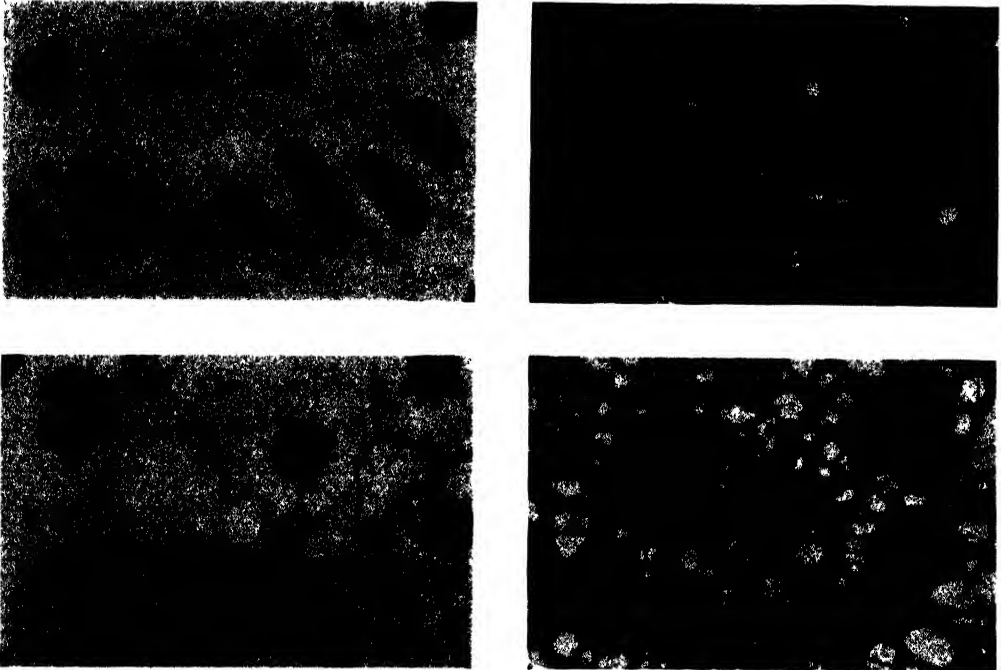
X chromosome and the Y chromosome of *Drosophila melanogaster*, are essentially inert genetically, it is tempting to draw a parallel and suggest that the temporary heterochromatization of the condensed chromosome provides a mechanism for the inhibition of genic activity during those periods when the chromosomes lie on the spindle. Verification must await further studies.

2. Degradation by ribonuclease of ribonucleoprotein, and the stainability with acidic dyes of the protein thus freed: The discovery that treatment of cells with ribonuclease increased the stainability of cytoplasm, nucleoli, and chromosomes with orange G prompted experiments to determine whether similar results might be obtained with other acidic dyes. Light green, fast green, and acid fuchsin proved equally effective. Of these the two green dyes seemed most satisfactory for cytological studies because of a quality of coloration which, when viewed microscopically, permitted a quantitative estimate of concentration, and because they could be combined effectively with orcein or the purple fuchsin color developed in the Feulgen reaction, to provide a contrasting color in the chromosomes. Moreover, the intensity of green could be enhanced by "mordanting" with alkali, following the procedure recommended by Semmens and Bhaduri, although, as the experiments with orange G have indicated clearly, the use of sodium carbonate (as recommended by Semmens and Bhaduri) or of sodium acetate, borate, or citrate (which we have found to be equally effective) is not essential to the formation of the color, but only serves to intensify it.

Thus if a section through the meristematic region of a root tip of *Allium cepa* fixed in Flemming's fluid is hydrolyzed in an aqueous solution of ribonuclease while an adjoining section, which serves

as a control, is left in water at the same pH and temperature, and if both are stained simultaneously with Feulgen followed by light green, differentiation may be carried out so that the unhydrolyzed section shows purple chromatin, pale green nucleoli, and colorless cytoplasm, whereas the ribonuclease-treated section shows brilliant green cytoplasm and nucleoli, with a greenish hue overshadowing the purple color of the chromosomes. If, however, the ribonuclease-digested section is subsequently treated with pepsin and then stained, no such intensification of the green color results. (See plate 1.) These experiments suggest that ribonuclease effects a degradation of ribonucleoprotein, resulting in the formation of a protein that stains intensely with acidic dyes. Since pepsin cannot digest histones, this protein is presumably of the nonhistone (tryptophane-containing) type. Moreover, its distribution within the cell parallels that of the tryptophane-containing material as determined by histochemical application of Bates' modification of the May-Rose test (Year Book No. 46). These observations show that at least part of the ribonucleic acid present in chromosomes, cytoplasm, and nucleoli is combined with tryptophane-containing proteins.

3. Degradation of ribonucleoprotein by pepsin, and stainability of the ribonucleic acid with basic dyes: With the knowledge in hand that ribonuclease may degrade ribonucleoprotein, it remained to be determined whether pepsin may also effect such separation. Under these conditions it would be expected that the liberated ribonucleic acid, unless it were lost from the cell, would stain with basic dyes, and that a method could be developed for determining by color values the difference between ribonucleoprotein and ribonucleic acid. Preliminary experiments have given



Degradation of ribonucleoprotein by ribonuclease to protein stainable with light green and digestible by pepsin.

Prints from Kodachrome film of sections of root tips of lily, all stained simultaneously in Feulgen followed by light green. Upper left, control; upper right, after digestion with ribonuclease; lower left, ribonuclease treatment followed by pepsin; lower right, control for pepsin treatment, ribonuclease followed by boiled pepsin.

hopeful results, and work on the method is in progress.

4. Action of other proteases: Pepsin digests "tryptophane-containing" proteins but not histones; trypsin and chymotrypsin digest both. The rate of hydrolysis of histones by chymotrypsin, however, is

trypsin. It is hoped that further modifications of our procedures will provide this much-needed information.

CHEMICAL STUDIES

Analysis of amphibian nuclei. Nuclei and chromosomes from ovarian amphibian

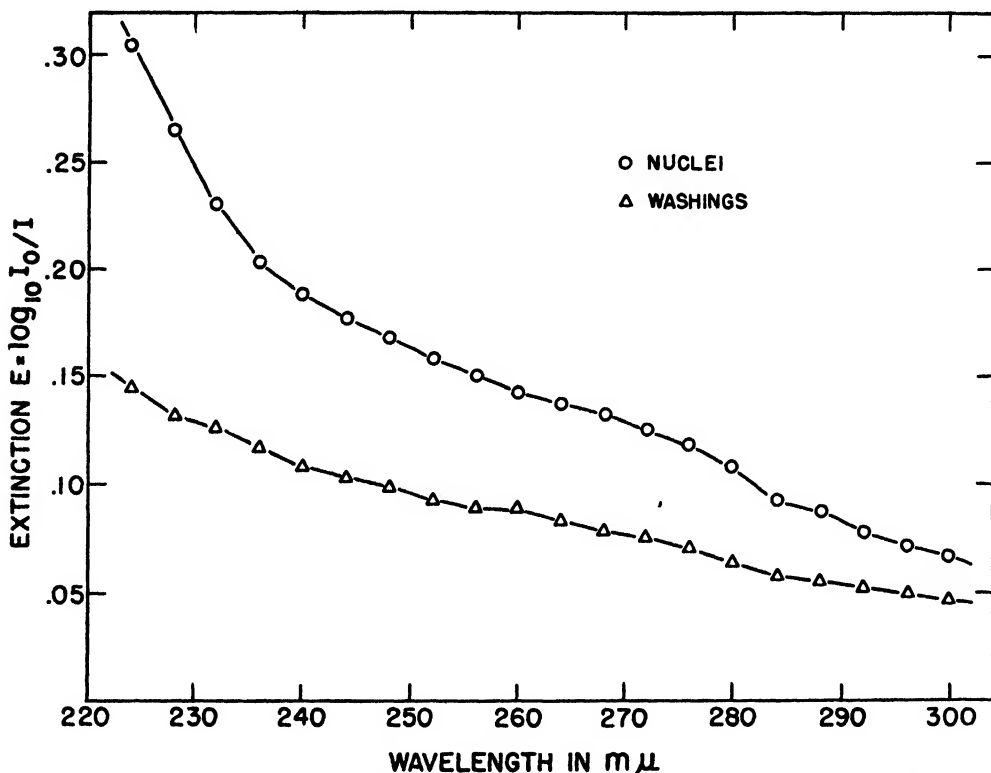


FIG. 1. The ultraviolet absorption spectra of the solutions resulting when (a) 50 large nuclei, isolated from ovarian eggs of *Triturus pyrrhogaster* by Duryee's technique, are homogenized in 2 ml. of calcium-free Ringer's solution plus 2 ml. of 0.2 N hydrochloric acid, and (b) 2 ml. of 0.2 N hydrochloric acid is mixed with the 2 ml. of calcium-free Ringer's solution in which the above nuclei had been washed after being freed of all apparent traces of cytoplasm and yolk. Control, 2 ml. of calcium-free Ringer's plus 2 ml. of 0.2 N hydrochloric acid. Length of absorbing column, 10 mm. The plotted values of E are observed readings of density $\log_{10} I_0/I$ of the solutions.

much slower than that by trypsin. Comparative studies of the rate of cellular digestion by these two enzymes should provide clues to the *in situ* location of histones. Unfortunately, our histochemical studies have so far yielded no indication of digestion by either trypsin or chymo-

eggs are extremely large, and can be isolated by microdissection techniques which eliminate some of the dangers inherent in the usual chemical procedures for the preparation of mammalian nuclei. From these standpoints, they appear to be excellent material for studying the changes

in chemical composition of nuclear constituents during the various stages of oögenesis. Studies of the total phosphorus content of whole eggs, enucleated eggs, and isolated nuclei of *Triturus pyrrhogaster* have shown, however, that the actual amount of nuclear material is very small

ings obtained in their isolation, that the dangers of cytoplasmic contamination are grave enough to indicate that, even if fully adequate micromethods were developed for the analysis of the various nuclear constituents, no unequivocal interpretation of the results could be obtained.

TABLE 1

INHIBITION OF RIBONUCLEASE BY DESOXYRIBONUCLEIC ACID: EFFECT OF VARYING THE CONCENTRATION OF DESOXYRIBONUCLEIC ACID AND OF RIBONUCLEASE, THE CONCENTRATION OF RIBONUCLEIC ACID BEING CONSTANT

Desoxyribonucleic acid (A).. Ribonucleic acid (B).....	10 ⁻³ MG. PER ML. DIGESTION MIXTURE										
	0	167	334	501	668	835	1002	1169	1336	1503	1670
	1670	1670	1670	1670	1670	1670	1670	1670	1670	1670	1670
A/B.....	Parts A per 100 parts B										
	0	10	20	30	40	50	60	70	80	90	100
Ribonuclease added..... Ribonuclease found.....	10 ⁻³ MG. PER ML. DIGESTION MIXTURE										
	120	120	120	120	120	120	120	120	120	120	120
	120	97	87	80	73	67	60	57	53	50	50
Inhibition.....	Per cent*										
	0	19	28	33	39	44	50	53	56	58	58
Ribonuclease added..... Ribonuclease found.....	10 ⁻³ MG. PER ML. DIGESTION MIXTURE										
	240	240	240	240	240	240	240	240	240	240	240
	240	190	173	153	147	133	120	117	103	100	97
Inhibition.....	Per cent*										
	0	21	28	36	39	45	50	51	57	58	60
Ribonuclease added..... Ribonuclease found.....	10 ⁻³ MG. PER ML. DIGESTION MIXTURE										
	360	360	360	360	360	360	360	360	360	360	360
	366	287	253	233	210	193	183	173	163	160	150
Inhibition.....	Per cent*										
	-2	20	30	35	42	46	49	52	55	56	58

*The percentage inhibitions noted here cannot be compared with those in table 2 because of differences in type and amounts of electrolytes present.

(<0.1 µg. of total phosphorus per nucleus) and represents, depending somewhat upon the stage of oögenesis, only about 2 per cent of the total material of the egg. Studies of the ultraviolet absorption spectra of these substances have led to the same conclusions. It is apparent from figure 1, which shows the absorption spectrum of dissolved homogenized nuclei as compared with that of the final wash-

Inhibition of ribonuclease by desoxyribonucleic acid. During our histological studies it was noted that cytoplasmic ribonucleic acid was more easily digested by ribonuclease than was the ribonucleic acid present in chromosomes. Since chromosomes are known to contain large amounts of desoxyribonucleic acid, whereas cytoplasm does not, it seemed possible that this nucleic acid was in some way

interfering with the action of ribonuclease. Test-tube experiments were therefore devised to test this possibility. It was found that under appropriate conditions as little as 1 part by weight of desoxyribonucleic acid, when added to 250 parts of ribonucleic acid, would greatly inhibit the action of ribonuclease. The amount of inhibition is dependent upon many factors. It is, for example, markedly altered by salts (being, in general, much greater the lower the salt concentration) and by pH. Since different ions appear to have different effects, however, and since varying the pH involves changing the concentrations of these ions, the latter effect may be illusory.

The inhibition has been studied by varying the concentrations of ribonuclease (enzyme), ribonucleic acid (substrate), and desoxyribonucleic acid (inhibitor) both alone and in relation to one another. At present only tentative conclusions about the mechanism of the inhibition can be drawn, partly because many variables are involved and partly because the studies are still incomplete. Certain mechanisms can be eliminated, however. If the inhibition were due to the formation of a nondissociable compound between enzyme and inhibitor, the amount of ribonuclease inhibited would be directly proportional to the concentration of desoxyribonucleic acid present and would be independent of the total concentration of ribonuclease. That this is not the case is seen in table 1, which shows the effect of varying the concentration of desoxyribonucleic acid and of ribonuclease, the concentration of ribonucleic acid being constant. Both the amount and the percentage of ribonuclease inhibited per unit weight of desoxyribonucleic acid decrease rapidly as the concentration of the latter is increased. The amount of ribonuclease inhibited by any one concentration of desoxyribonucleic acid appears to be, with-

in the limits of experimental error, directly proportional to the concentration of ribonuclease used, the percentage of inhibition being practically constant. This fact eliminates the possibility that the inhibition is due to a reversible combination of enzyme and inhibitor, nondissociable by the substrate, for in that case the smaller the quantity of ribonuclease, the greater should be the percentage of inactivation. This possibility must also be rejected on the basis of the results obtained from experiments in which the concentration of ribonucleic acid was varied. If the substrate played no role in the inhibition, the activity of the enzyme would be equally reduced at all substrate concentrations. The data in table 2 show, however, that for any one concentration of ribonuclease and desoxyribonucleic acid, an increase in the concentration of ribonucleic acid brings about a marked decrease in the amount of inhibition, and that the percentage inhibition appears to be a function of the ratio of the concentration of ribonucleic to desoxyribonucleic acid. These facts suggest either that the inhibitor and substrate unite to form a complex not digestible by the enzyme, or, more probably, that there is competition by the two nucleic acids for the enzyme. It is hoped that further experimentation will clarify the situation.

Solubility of proteins in Carnoy's alcohol-acetic fixative. One of the first requirements of a good cytological fixative is that it should preserve intact all the cellular components to be studied. Carnoy's alcohol-acetic acid mixture has been widely used in obtaining quantitative data on the amount of desoxyribonucleic acid, ribonucleic acid, histone, and tryptophane-containing protein present in cells. The finding, reported by McDonald in Year Book No. 44 (1944-1945), that certain proteins such as serum albumin are soluble in acidified alcohol has led to a study of

the components found in Carnoy's fluid after the fixed tissues have been removed. Preliminary experiments have shown that crystalline serum albumin is soluble in the alcohol-acetic acid mixture, and that some protein is lost from specimens (root tips) that have been fixed in this fluid.

now been tested by studies of chromosome rearrangements and dominant and recessive lethals. Near infrared radiation in itself does not induce gene mutations or chromosome breaks. No modification of the over-all frequency of induced dominant or recessive lethals was obtained when

TABLE 2

INHIBITION OF RIBONUCLEASE BY DESOXYRIBONUCLEIC ACID: EFFECT OF VARYING THE CONCENTRATION OF RIBONUCLEIC ACID, THE CONCENTRATION OF DESOXYRIBONUCLEIC ACID BEING CONSTANT

	10 ⁻⁵ MG. PER ML. DIGESTION MIXTURE									
	100	100	100	100	100	200	200	200	200	200
Desoxyribonucleic acid (A).....	625	1250	1875	2500	3125	625	1250	1875	2500	3125
Ribonucleic acid (B).....	Parts A per 1000 parts B									
A/B.....	160	80	53	40	32	320	160	107	80	64
	10 ⁻⁶ MG. PER ML. DIGESTION MIXTURE									
	200	200	200	200	200	200	200	200	200	200
Ribonuclease added.....	52	88	104	118	129	33	50	68	84	99
Ribonuclease found.....	Per cent*									
Inhibition.....	74	56	48	41	36	84	75	66	58	51
	10 ⁻⁶ MG. PER ML. DIGESTION MIXTURE									
	100	100	100	100	100	100	100	100	100	100
Ribonuclease added.....	27	42	52	60	66	18	28	36	44	47
Ribonuclease found.....	Per cent*									
Inhibition.....	73	58	48	40	34	82	72	64	56	53

*The percentage inhibitions noted here cannot be compared with those in table 1 because of differences in type and amounts of electrolytes present.

This fact must therefore be considered in interpreting quantitative data on the protein content of alcohol-acetic acid-fixed tissues. The type of protein(s) lost and its *in situ* location has not yet been determined.

FURTHER STUDIES UTILIZING INFRARED RADIATION

Combination treatments. The effect of the near infrared portion of the spectrum (wave lengths centering around 10,000 Å) in modifying the action of ionizing radiations on spermatozoa of *Drosophila* has

near infrared radiation was used in combination with X-rays. However, exposure of spermatozoa to near infrared radiation before treatment with X-rays significantly increased the frequency of viable types of rearrangement as compared with the frequency in controls receiving only the X-rays. (See Year Books No. 41 [1941-1942] *et seq.*) We have during the past year confirmed our previous observation that treatment of mature spermatozoa with near infrared after X-radiation does not increase the frequency of rearrangement.

Our more recent posttreatment experi-

ments have involved exposure of females to near infrared radiation after their insemination by X-ray-treated males. This procedure obviates the effect of posttreatment on the progress of spermatogenesis, detected in earlier studies. Larvae developing from eggs laid after the infrared treatment provided the salivary-gland-chromosome preparations utilized in determination of break frequency and types of induced changes. The data obtained are summarized in table 3.

chemical, we utilized genetical rather than cytological methods of analysis in our primary assay. Males were exposed to vapors of the chemical in the treatment chamber designed by Demerec, then mated with females carrying the dominant markers Curly and Plum in their second chromosomes, and Hairless and Stubble in their third chromosomes. The progeny of this cross were then tested individually for evidence of reciprocal translocation between the second and third chromosomes.

TABLE 3
EFFECT OF POSTTREATMENT WITH NEAR INFRARED RADIATION

Treatment	Total sperm tested	No. rearrangements	Per cent sperm showing rearrangements	No. lethals	Breaks per 100 sperm
♂♂ ca. 5000 r X-rays; inseminated ♀♀ 48 hrs. at 18° C.	263	125	47.5 ± 3.1	335	127.4
♂♂ ca. 5000 r X-rays; inseminated ♀♀ 48 hrs. of near infrared.	315	158	50.2 ± 2.8	427	135.5
Diff./S.E. = 2.7/4.2 = 0.64					

It is thus apparent that although the frequency of chromosomal rearrangement induced by X-ray treatment of the mature spermatozoa of *Drosophila* can be modified by near infrared radiation used before the X-rays, it cannot be when the sequence is reversed. The question arises whether similar results might be obtained if near infrared were used in combination with other agents that induce chromosome breakage. We have accordingly carried out a series of experiments with the chemical mutagen, nitrogen mustard—methylbis(betachloroethyl)amine hydrochloride. Mr. Harvey Rothberg, of Princeton University, assisted in this project.

Because of the low frequency of rearrangement obtained after exposure of *Drosophila* males to nonlethal doses of the

Pretreatment of males with near infrared radiation provided a significantly higher frequency than that obtained in the controls, which were kept at 18° C. during the 24 hours their brothers were exposed to the infrared beam, but were exposed to nitrogen mustard at the same time and in the same container as the others. On the other hand, the frequency among spermatozoa exposed to near infrared radiation after the nitrogen mustard was not significantly different from that for their controls. These results in the pretreatment and posttreatment experiments parallel those obtained with X-rays, and confirm our findings concerning the efficacy of pretreatment with near infrared radiation in increasing the yield of induced chromosomal rearrangements.

Other similarities between the action of nitrogen mustard and that of ionizing radiations concern the random distribution of breaks among chromosomes and along the chromosomes. To secure information about the distribution of nitrogen mustard-induced breaks, each of the genetically determined translocations was subjected to further analysis by inspection of salivary-gland chromosomes. The data obtained indicate that breaks are distributed along the chromosomes in proportion to length represented in mitotic stages, irrespective of division into euchromatic or heterochromatic regions. This finding suggests that the basic pattern of organization of the chromosome is similar throughout its length.

Although it is tempting to utilize the similarities in patterns of response of germinal materials as evidence of similarity of mode of action of nitrogen mustard and ionizing radiations, there are some striking differences in the response to the two mutagenic agents. One such difference has been revealed in the study here reported. Spermatozoa transferred in copulation within the first 12 to 14 days after an X-ray treatment provide a fairly constant and uniform frequency of types of induced aberrations, such as translocations. Among spermatozoa transferred after this period, there is a marked decrease in the frequency of aberration; and this has been attributed to the utilization in copulation of cells that were not mature at the time of the X-ray treatment. No such decrease occurs among spermatozoa of males exposed to nitrogen mustard; there is on the contrary a significant increase with passage of time for the duration of the period that fertility continues (about 21 days in these experiments). The cytological studies have shown that the increase is not attributable to clusters of similar translocations originating at early

stages of spermatogenesis. This finding raises several questions concerning the action of the mutagenic products of nitrogen mustard treatment, particularly with respect to selective action on the individual chromonemata of the chromosomes of the spermatozoa.

Qualitative differences among breaks. The survey that we have conducted to determine the influence of near infrared radiation on induced gene and chromosome alterations in *Drosophila* indicates that the sensitizing action of this portion of the spectrum is restricted to the control of processes involved in the formation of chromosomal rearrangements. No general increase in number of all types of breaks is involved, for pretreatment does not increase the frequency of the single-break type of dominant lethal. These facts suggest that induced breaks fall into qualitatively different classes, since the subsequent behavior of some breakage ends was modified by conditions created as a consequence of pretreatment, whereas that of others was not. The question arises whether such qualitative differences may be ascribed to variations in the time at which the types of aberrations are produced. Although the experiments utilizing near infrared radiation do not in themselves answer this question, they provide clues that can be appraised in the light of other evidence. They suggest that the single-break type of dominant-lethal aberration of *Drosophila* may be produced by end-to-end union of sister chromatids at the time of irradiation or shortly thereafter. Available evidence also suggests that some restitution of potential breaks may take place before the time of recombination, which presumably occurs in *Drosophila* after the spermatozoon has penetrated the egg in fertilization (Year Book No. 39 [1939-1940]). Thus, by means of the various combination-treat-

ment experiments we are gradually accumulating the information essential to a critical analysis of the sequence of events in the process of structural rearrangement of chromosomes.

Potentialities of near infrared radiation. Of the various types of supplementary treatment utilized, near infrared radiation has proved most serviceable in analysis of the cycle of breakage and recombination. This agent modifies the processes that lead to structural rearrangement, but not those responsible for gene mutation. Accordingly, near infrared radiation may serve as a useful diagnostic tool, if applied to organisms in which cytological analysis is not feasible, for distinguishing between genetic changes that are attributable to chromosomal alterations and those that are not. The potentialities of this portion of the spectrum for further cytogenetic studies lie in the prospect of securing intense monochromatic radiation of known physical properties. In order that such methods may be utilized more efficiently than in the laborious procedures involved in cytological analysis of salivary-gland chromosomes for induced rearrangements, we have turned our attention in recent months to other methods of quantitative assay of the effects of the combination treatments. Experiments are now being carried on to determine the influence of near infrared radiation (1) on the process of crossing over, which involves spontaneous exchange between chromatids of homologous chromosomes, (2) on the inactivation by X-rays of solutions of purified crystalline enzymes, such as trypsin and ribonuclease, and (3) on the frequency of induced mutation in strains of the bacterium *Escherichia coli*. Miss Helen Cuneo, of Antioch College, has assisted in the work on *E. coli*. The findings will be reported later.

NONHOMOLOGOUS ASSOCIATION OF SALIVARY-GLAND CHROMOSOMES

Analysis of patterns of nonspecific pairing among the bands of the salivary-gland chromosomes of *Drosophila melanogaster* has been continued. Additional observations have been made by Dr. Marcia Kelman Iddles, working in co-operation with Kaufmann. Identification of the bands involved in more than 400 cases of nonspecific association has provided the type of information portrayed in figure 2, which shows the observed cases of pairing within the X chromosome, and between the X and the second and third chromosomes. Comparable data are available for each limb of these autosomes.

In figure 2 the map of the X chromosome has been arranged in the form of a crescent. Within the curve, connecting lines indicate the observed cases of association between different parts of the X. Outside the curve the numbers of the bands of the X chromosome are represented, and beyond these, connected by straight lines, are the numbers of the bands in the autosomes with which the X has been found to be associated. As an example, starting at the tip of the X, we find that the complex 1A1,2,3,4 has been observed to pair with bands 3B1,2 in the X, and also with 61A1, which is in the left limb of the third chromosome, and 100F1,2,3,4,5, which is at the tip of the right limb of the third chromosome. The latter pattern was observed twice, and for that reason the numeral 2 is shown in parentheses.

Some patterns of association occur more frequently, as is seen in divisions 11 and 12. Thus, 11A6,7 was observed in association with 11A8,9 in 18 cases. These adjacent bands frequently adhere at one end but separate widely at the other to present

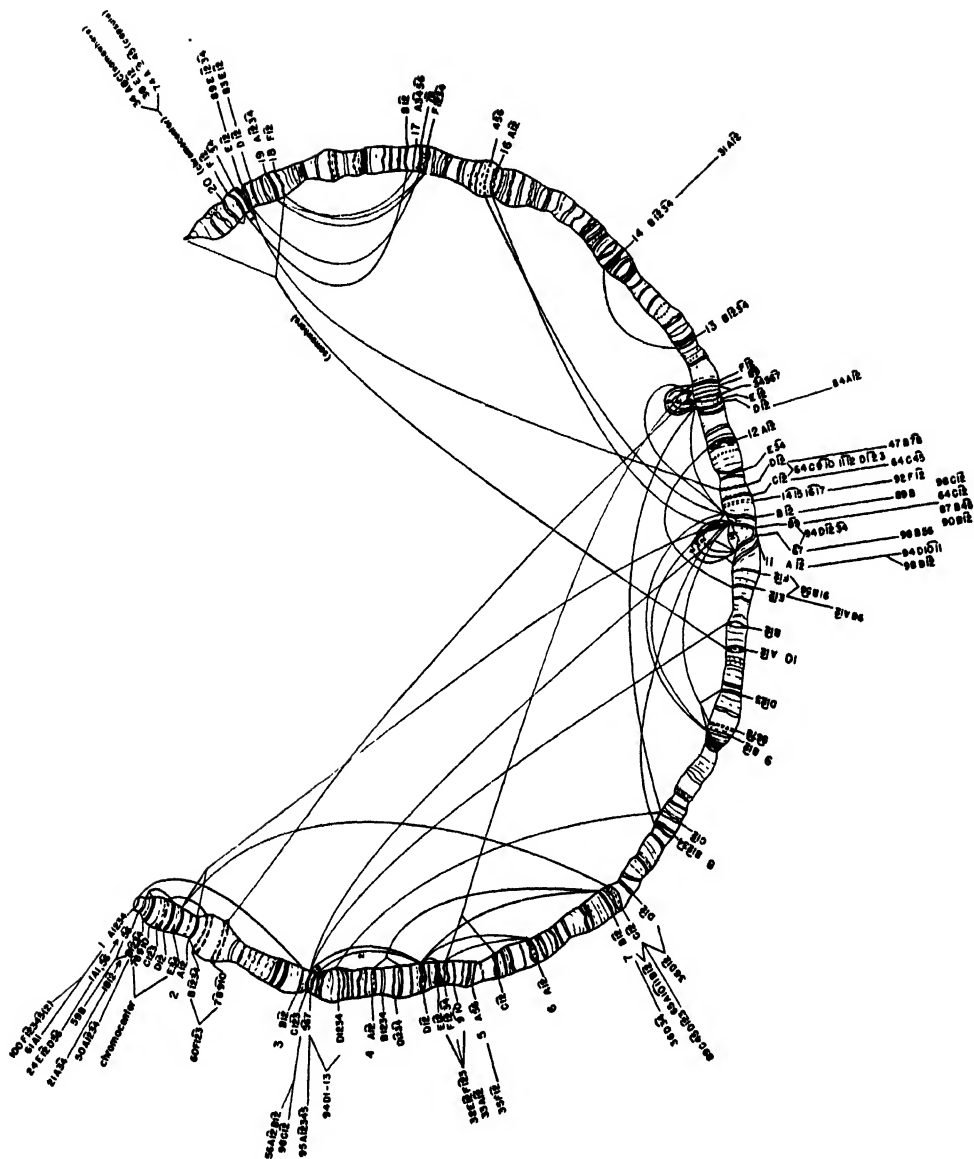


FIG. 2. Patterns of nonhomologous association of the bands of the X chromosome of *Drosophila*

a V-shaped or linear pattern, so that the chromosome appears broken open. Proximity favors this pattern of association, but these bands also have been found to establish connections with other regions of the X (such as 2A_{1,2} and 3C_{1,2,3}), or of the autosomes (such as 64C_{1,2}; 87B_{4,5}; 94D_{1,2,3,4}; etc.). Since several lines of evidence have indicated that subdivisions 11A and 12DE contain heterochromatin, associations between these subdivisions and others may provide a guide to the location of islets of intercalary heterochromatin that have not been detected by other methods of analysis (such as the break-frequency method outlined in Year Book No. 45 [1945-1946]). Final conclusions must await the integration of various lines of experimental evidence. For the moment, the data provided by these observations of nonhomologous association are serving as a guide in our studies of the action of various enzymes on the different regions of the salivary-gland chromosome.

Apart from the heterochromatic regions, there are various duplicated euchromatic

regions distributed among the chromosomes. Frequently these regions lie side by side, and a method by which such linear duplication may occur in the course of phylogeny was established experimentally several years ago by Kaufmann and Bate (see Year Book No. 37 [1937-1938]). But it is also possible that such repeats may have become more widely separated in the process of evolution of the chromosome. Thus, patterns of association of nonhomologous regions must be examined with a view to locating duplicated euchromatic regions. An effort was accordingly made to determine whether the loci of any genes having similar phenotypic effects are associated with regions in which the bands undergo nonhomologous association. Although *Drosophila melanogaster* is genetically the best known of all organisms, there is a dearth of information concerning the cytological localization of specific genes. For this reason no correlation of the type sought has been immediately apparent, but analysis of the data is being continued.

MUTABLE LOCI IN MAIZE

BARBARA MCCLINTOCK

Previous reports have stated that a number of unstable loci have recently arisen in the maize cultures. In a particular cell of a plant, a normal "wild-type" locus becomes altered; the normal, dominant expression of this locus changes and gives rise to a recessive expression (or, in several cases, a recessive locus becomes unstable and mutates toward a dominant expression). This expression of the locus need not be permanent. In some descendent cells, a second change may occur within the locus that results in the restoring of the capacity of the locus to express the dominant phenotype or brings about an intermediate expression between full recessive and full

dominant. In the latter case, a third alteration may occur in some descendent cells that steps up the phenotypic expression toward the full dominant or reduces it toward the full recessive.

When a locus changes from a stable to an unstable state, recognition of the occurrence depends upon several factors. A clear-cut phenotypic expression of the changed locus is essential. This limits ease of recognition to those gene loci associated with the production of some obvious plant constituent, such as the chlorophyll pigments or other plant pigments, or to contrasting morphological characters striking enough to allow the contrasts to be de-

tected in single small sectors of some part of the plant. The presence of a changed locus that gives a recessive expression often may remain hidden in a heterozygous state until a self-pollination or some special cross is made that allows its presence to be uncovered. No attempt has been made to detect the presence of these hidden changed loci, and thus the frequency of occurrence of such changes is not known. The unstable loci that have been discovered have all appeared in the progeny of crosses designed for other purposes. It is suspected that the number of mutable loci still undetected may be quite great, with numerous independent occurrences of unstabilization of the same locus. The detected unstable loci include many previously unknown to maize geneticists. Some well known loci in maize have also become unstable in these cultures. These include two independently arising unstable *yg* loci (yellow-green chlorophyll), four independently arising unstable *c* loci (*c*, colorless aleurone; *C*, colored aleurone), three independently arising unstable *wx* (waxy) loci (*wx*, starch staining red with iodine; *Wx*, starch staining blue with iodine), and one unstable *a₂* locus (anthocyanin in aleurone and plant). With the exception of the two *yg* loci, all these unstable loci have originated by a change in a normal, previously quite stable locus showing dominant expression. In the case of unstable *yg*, the recessive *yg* locus mutates to form chlorophyll of a much darker color than the normal *yg* locus expresses.

These mutable loci fall into two distinct classes: (1) those that require the presence of a second locus, the activator locus (*Ac*), for instability to be expressed and maintained, and (2) those that do not require such a second locus for instability to be expressed. The *Ac* locus itself is unstable

and resembles in this respect the second class of unstable gene loci.

Two recognizable subdivisions of the mutation process are shown by all the unstable loci: (1) control of the time and the apparent frequency of mutations in a tissue by a factor capable of changing during a mitosis so that altered rates of mutation will be expressed in descendent cells following such a change; and (2) the subsequent change at the locus that occurs in a particular cell and gives rise immediately to recognizable altered phenotypic expression in this cell and its descendent cells. The term "state of a locus" has been used to distinguish the differing potentialities of a mutable locus for expressing visible mutations in descendent cells. The mutations giving rise to changes in state of a locus are readily recognized by the altered rates of the second type of mutation that follow from such an event. In the second class of mutable loci—those controlled by *Ac*—the time and apparent frequency of mutations of the affected locus are controlled by the states of both loci: the locus controlled by *Ac* and the *Ac* locus itself.

During the past year, attention has been directed mainly to those loci that require the *Ac* locus for continued expression of instability. All these loci that are known are in the short arm of chromosome 9, possibly because the genetic methods being used allow certain mutable loci in this arm to be readily detected. Those receiving particular study include the *Ds* (Dissociation) locus, mentioned in previous reports, two independently arising mutable *c* loci, and two independently arising *wx* loci. In each case, the mutable recessive state of the previously stable dominant locus arose in a particular cell of a plant that also possessed an *Ac* locus. The origin of this potent *Ac* locus is quite unknown. It has been traced to a culture

grown in the summer of 1942, but its presence in still earlier cultures cannot be traced. It cannot be stated that *Ac* induces the initial state of instability of a locus, although a causal connection may be suspected.

None of these *Ac*-controlled mutable loci will show any type of instability if the *Ac* locus is absent from the nucleus. As soon as *Ac* is introduced into a nucleus having such a susceptible locus, instability is resumed. Mutations at the susceptible locus sometimes may occur in the initial nucleus following the introduction of *Ac*. To illustrate the control of mutability of a locus by *Ac*, the mutable c^{m-1} locus may be used. This locus has been given the symbol c^{m-1} because it was the first of the several mutable *c* loci isolated; the normal *c* locus, used for many years in genetic experiments, is designated c^s because it is a stable locus not mutating under the influence of *Ac*. The behavior of this unstable recessive *c* locus in the designated constitutions is indicated below:

- c^{m-1} *ac*: no mutations; aleurone layer colorless
- c^s *ac*: no mutations; aleurone layer colorless
- c^s *Ac*: no mutations; aleurone layer colorless
- c^{m-1} *Ac*: mutations to *C* occur; aleurone layer variegated for color

It should be emphasized that, with similar constitutions, the same type of responses will be registered by any of the other loci that are activated by *Ac*.

NATURE OF THE *Ac* ACTION

The Ds locus. The *Ds* locus was discovered before any of the other *Ac*-controlled unstable loci. *Ds* is located at the position demarking the proximal third of the short arm of chromosome 9, between one and two crossover units to the right

(toward the centromere) of the *Wx* locus. The normal, nonmutating locus has been designated *ds*. An observable *Ds* mutation arises as the consequence of breakage of two sister chromatids within the *Ds* locus in each case. This breakage is followed by fusions of broken ends to give rise to a U-shaped acentric fragment composed of the two sister segments of the distal two-thirds of the short arm, and a U-shaped dicentric chromosome composed of the proximal third of the short arm, the centromere, and all of the long arm of each of the two sister chromatids. Nearly all the known loci of this chromosome are carried in the distal two-thirds of the short arm of chromosome 9 and are thus included in the acentric fragment after a *Ds* mutation. The genetic methods of observing the time and frequency of the *Ds* mutations in various parts of the plant have been explained in previous reports (Year Books Nos. 45 [1945-1946] and 46 [1946-1947]).

The *Ac* locus acts upon *Ds*, or any other locus susceptible to it, usually quite late in the development of any of the sporophytic tissues. In contrast, action of *Ac* on susceptible loci may be apparent at all stages in the development of the endosperm tissues. In this respect, the endosperm behaves like an extension of the sporophytic tissues.

As stated above, the detectable *Ds* mutations are associated with breaks at this locus that are followed by 2-by-2 fusions of those broken ends of sister chromatids that lie adjacent to one another. This type of fusion following breakage is not the only one that could occur. Restitutions could take place, re-establishing the previous organization of the chromatids; or crisscross fusions of broken ends could occur, simulating crossing over but involving sister chromatids in somatic nuclei. It is not known whether or not the latter

types of fusion do occur with expected frequencies at the *Ds* locus. The evidence suggests that they may occur, at least occasionally. It is known that breaks at the *Ds* locus may be followed directly or indirectly by fusions of broken ends other than those giving rise to the U-shaped dicentric chromatids. This is shown by the fusions that occur after an occasional coincidental break in another part of the chromosome. It is possible then for a broken end associated with a *Ds* mutation to fuse with one of these other newly broken ends. This gives rise to a gross chromosomal aberration that can be analyzed. Such chromosomal aberrations have been observed in individual cells or clusters of cells in the examined sporocytes of the *Ds Ac* plants. Several of these translocations have been found in individual plants, and have received further study. These aberrations have been useful in interpreting occasional inconsistencies in the behavior and the location of the *Ds* and the *Ac* loci. It is now known that the *Ds* locus may change its position in the chromosome after such coincidental breaks have occurred. One very clear case has been analyzed, and, through appropriate selection of crossover chromatids, strains having morphologically normal chromosomes 9 have been obtained. As a consequence of this aberration, the *Ds* locus in these strains has been shifted from a position a few units to the right of *Wx* to a position between *I* and *Sh*. This is a very favorable position for showing the nature of the *Ds* mutation process. When the usual type of *Ds* mutation occurs at the locus in this new position, a U-shaped dicentric chromosome is formed. At the succeeding anaphase a chromatin bridge is produced, which subsequently breaks. The breakage-fusion-bridge cycle is thus initiated and can be genetically expressed in succeeding nuclear divisions. This is

because the loci of *Sh*, *Bz*, and *Wx* now lie between *Ds* and the centromere. With appropriate genic constitutions of chromosome 9, detection of the breakage-fusion-bridge cycle is certain after a *Ds* mutation has occurred in this new location. These observations have been made, and the genetic analysis completely confirms the cytological analysis of the nature of the *Ds* mutation process.

The inheritance of Ac as a separate locus. *Ac* behaves in inheritance as a single independent locus. Tests for the presence of *Ac* in F_2 progenies of *ds ds Ac ac* plants have given ratios of 1 *Ac Ac* to 2 *Ac ac* to 1 *ac ac*; backcrosses of *Ac ac* plants by *ac ac* plants have given ratios of 1 *Ac ac* to 1 *ac ac* plant. Crosses of *Ac Ac* plants by *ac ac* plants have given progenies all of which were found to be *Ac ac*. In the many crosses that have been made with plants that were heterozygous for both *Ds* and *Ac*, independent inheritance of the *Ac* and *Ds* loci was clearly established, except in three independently arising cases. In these three cases, *Ac* was obviously linked with *Ds* and could be located 6 to 20 crossover units to the right of it. Such linkages were maintained in later tests of the progeny of one of these plants, when both the crossovers and the noncrossover chromatids were tested. Similar tests are now under way for the other two cases. Cytological examination of heterozygous plants in each of the three cases showed no observable chromosomal abnormalities. Chromomere matching between synapsed homologues was perfect for all of chromosome 9.

Either of two possibilities may explain this change in genetic location of *Ac*. First, *Ac* may be located toward the end of the long arm of chromosome 9 and normally show no genetic linkage with *Ds* because the crossover distance is too great. In the case of *Ds-Ac* linkage, a newly

arising crossover modifier, not associated with a gross chromosomal abnormality, would need to be invoked to explain the observations. This modifier would need to be closely linked with *Ac*, as it has not been removed in the crossover tests so far made. Second, the *Ac* locus may have been removed from its former position and inserted into a new position in chromosome 9 in a manner similar to that observed for the transposition of the *Ds* locus, described above. Because *Ac* induces breaks at specific loci and gives evidence of undergoing a specific breakage process itself, this latter explanation is not improbable. Tests are now under way to determine whether or not either of these alternatives can apply.

The effects of dosage of Ac. Studies of the effects of various dosages of *Ac* have shown that the time and apparent frequency of *Ac*-controlled mutations is in large measure a function of dosage of *Ac*. This applies both to the sporophytic tissues and to the endosperm tissues. The endosperm tissue is $3n$. Here, one, two, or three doses of *Ac* can be obtained and observations made of the effects of each of these dosages on any of the *Ac*-controlled mutable loci. All *Ac*-controlled mutable loci respond alike to the various *Ac* dosages. The description given below of responses of the *Ds* locus in endosperm tissues can be applied equally well to the responses of any of the other *Ac*-controlled mutable loci.

In *Ds ds ds* kernels, the mutation rates of the single *Ds* locus have been compared in *Ac ac ac*, *Ac Ac ac*, and *Ac Ac Ac* endosperm constitutions. One dose of *Ac* allows mutations to occur relatively early; considerable irregularity in both time and frequency of mutations is apparent, for the endosperm is often divided into various larger or smaller sectors each showing its own special rate of mutation. This irregu-

larity is only apparent, as subsequent evidence will show. Two doses of *Ac* delay the timing of *Ds* mutations so that they occur relatively late in the development of the endosperm. The frequency may be very high, however. When three doses of *Ac* are present, only relatively few, very late-occurring *Ds* mutations are recognized in these kernels. Three doses of *Ac* appear to decrease the frequency of *Ds* mutations, but this appearance is probably deceptive. It is more likely that the increased dosage of *Ac* so delays the timing of *Ds* mutations that only the very earliest-occurring of these can possibly show in the endosperm, and that if the endosperm cells had continued to divide, very large numbers of *Ds* mutations would have appeared. In other words, the tissues mature before these potential *Ds* mutations can be expressed. The validity of this interpretation will be apparent when the various states of the *Ac* locus are defined and their dosage responses compared. It may be concluded, then, that the absolute dosage of *Ac* is one main factor controlling the time and apparent frequency of mutations: the higher the *Ac* dosage, the later the occurrence of *Ds* mutations.

The frequency of *Ds* mutations is complicated to analyze because of three factors: (1) the maturing of tissues with high dosages of *Ac* before *Ds* mutations can be expressed; (2) changes in state of a single *Ac* locus, resulting in changes in response of the *Ds* locus that resemble either an increase or a decrease in the dosage of *Ac*; and (3) changes in state of the *Ds* locus, resulting in changes in response to *Ac* dosages. Factor (1) has been briefly considered above. Factor (2) will be considered next, and factor (3) will receive brief consideration later.

Changes in state of the Ac locus. As mentioned above, in the endosperms of all

kernels having the constitution *Ds ds ds* and *Ac ac ac* there is great variability in the time and frequency of *Ds* mutations within a single kernel. Various numbers of relatively large or small sectors may be present, each showing a special time and frequency of *Ds* mutations, or showing no *Ds* mutations at all. In most of these kernels, a few relatively early *Ds* mutations are observed. Several male parents of the constitution *Ds ds Ac Ac*, when crossed to *ds ds ac ac* female plants, produced kernels with unusually favorable sectors of the above-mentioned types. In over 90 per cent of the *Ds*-carrying kernels, sectoring occurred early in the development of the endosperm. Each sector showed a very distinct type of mutation pattern. The sectors in two of the most common classes of kernel will be described for the purpose of illustration. In one class, the kernel was composed of four very distinct sectors. In one sector, no *Ds* mutations or only a few very late mutations occurred; in the second sector, the *Ds* mutations occurred late but relatively frequently and were uniformly distributed within the sector; in the third sector, the *Ds* mutations occurred at a time midway in development of the endosperm and at a medium rate; there were fewer mutations per area than in the second sector, and they occurred earlier. The fourth sector arose from a nucleus that had undergone a *Ds* mutation; no further *Ds* mutations could be registered in this sector. The second most frequent type of kernel was composed of only three sectors, similar to the first, second, and fourth sectors of the above-mentioned type. It had no sector of medium-timed mutations. Nearly all the kernels resulting from these crosses could be classified on the basis of the presence of combinations of two, three, or all four of these distinct types of sectors. The origin of a sector,

in each case, could be traced to a nucleus arising from the earliest divisions in the endosperm. The ratio of kernels with specific combinations of sectors was surprisingly constant. Some combinations occurred with high frequencies, others with relatively low frequencies; and some combinations were not found at all.

It is obvious that something occurs in the early divisions of the endosperm that results in nuclei with differing potentialities for expressing the time and frequency of *Ds* mutations. Immediately upon examination of these kernels, one is impressed with the resemblance of the mutation patterns in the various sectors to the patterns that have been obtained by combining various dosages of *Ac*. Here, however, only one *Ac* locus was introduced into the primary endosperm nucleus. Obvious questions present themselves: Do these sectors arise as the consequence of some abnormality that results in segregations of various dosages of *Ac* in these early nuclei? If so, what abnormalities occur and what is the segregation mechanism? Neither nondisjunction of chromatids containing *Ac*, nor transposition of the *Ac* locus to another position in the chromosome set followed by mitotic segregation, satisfactorily explains the observed ratios of the various types of kernels. A satisfactory fit is obtained if it is assumed that (1) the *Ac* locus is composed of a number of identical and probably linearly arranged units, and (2) changes in the number of units can take place at the locus during or after chromosome reduplication such that one chromatid gains units that the sister chromatid loses. If the *Ac*-carrying chromosome is double at the time of entrance into the primary endosperm nucleus, such an event involving one of the two chromatids during the subsequent division could result in four nuclei, two with unaltered units within the *Ac* locus

and two with altered units within the *Ac* locus (one of these having an increased number of units and the other a decreased number). The four resulting nuclei would not be alike with respect to dosage of units within the single *Ac* locus. If the number of units within the *Ac* locus controls the time and frequency of *Ds* mutations in a manner comparable to known dosages of whole *Ac* loci, the time and frequency of *Ds* mutations in the various sectors could be explained. If such a change in units at an *Ac* locus is accompanied by a *Ds* mutation and involves one of the four chromatids carrying *Ds*, the kernels that have one sector produced as the consequence of a *Ds* mutation could be accounted for. This interpretation is subject to several kinds of test. It should be possible to predict the *Ac* type of control in those sectors that have undergone an early *Ds* mutation. Special genetic methods for detecting the *Ac* action in these sectors have been devised. Preliminary results from one test indicate a good fit with expectation; the most favorable tests should be available in the summer's harvest.

Other evidence supports the assumption that the *Ac* locus is composed of a number of identical units and that these units can increase or decrease as the consequence of a somatic mutation within the *Ac* locus. This considers what has been called the "state of the locus." The various states of the *Ac* locus are recognizable by the time and frequency of the mutations it produces when combined with one particular *Ds* locus. This particular *Ds* locus must have a constant state. A *Ds* locus can be maintained in a constant state by keeping it isolated in an *ac ac* plant; for without *Ac* the *Ds* locus is quite stable and remains unchanged. *Ac* loci have been isolated that produce in *Ac ac ac* constitutions relatively late *Ds* mutations.

When such late mutations result from a single dose of *Ac*, the *Ac* locus is considered to be in a high state. Other *Ac* loci have been isolated that in single doses induce many early mutations. These are called low-state *Ac* loci. In an *Ac Ac ac* constitution (two doses) a low-state *Ac* locus may give a mutation pattern resembling that obtained from a high-state *Ac ac ac* (one dose) constitution. Again, the *Ac Ac Ac* (three doses) constitution of a low-state *Ac* locus may give mutation patterns resembling those produced by a high-state *Ac Ac ac* (two doses) constitution. The effects produced by three doses of a high-state *Ac* locus are most instructive, because the delay in timing of mutations with this particular state and dosage of *Ac* is so great as to result in no visible mutations, or only an occasional one. The tissues have ceased dividing before the potential mutations can occur. The variously exhibited states of individually selected *Ac* loci and the effects produced by each in one, two, and three doses, respectively, give added support to the interpretation that the state of any one *Ac* locus is an expression of the number of reduplicate units present within this locus. It is obvious that the *Ac* locus itself is mutable, for *Ac* loci with different states can be recognized and isolated in the progeny of a cross of *Ac ac* by *ac ac*. It is suspected that what can be seen in the sectorial kernels described above are mutations of the *Ac* locus.

THE MUTABLE *c* LOCI

Mutable c^{m-1}. One of the two mutable *c* loci being studied originated from the cross of a female plant homozygous for *c sh ds ac* by a male plant homozygous for *C Sh Ds* and heterozygous for *Ac (Ac ac)*. The same male plant was used in making a number of similar crosses. On one of the

resulting ears, a single aberrant kernel was observed. Instead of showing either a complete *C* color (*C Ds/c ds/c ds, ac ac ac* constitution) or a variegation pattern composed of colored aleurone with colorless sectors, owing to *Ds* mutations (*C Ds/c ds/c ds, Ac ac ac* constitution), this kernel was obviously composed of colorless aleurone in which colored areas were present. In pattern of variegation, it was the reverse of expectation: a *c* locus appeared to be mutating to *C*. This kernel was removed, a plant was grown from it, and various crosses were made to determine the nature of the altered expression of aleurone color. Appropriate genetic analysis indicated that a mutable *c* locus was present which had arisen from the normal *C* locus present in the male parent. This *C* locus had changed to a mutable *c* locus capable of mutating back to the original *C*. This mutable *c* locus is *Ac*-controlled and responds in precisely the same manner as the *Ds* locus to various doses of *Ac* and to various states and observable changes in state of the *Ac* locus. With respect to *Ac*, the responses of the two loci are amazingly alike. *Ds* mutations, however, are known to be the result of some mechanism leading to chromosomal breakage and fusion, whereas the mutations at the *c* locus appear to involve quite a different mechanism, leading to changes in expression of the locus from recessive to dominant.

Similarities in response of *Ds* and c^{m-1} to *Ac*, together with the known breakage mechanism at the *Ds* locus and also the obvious changes in state of the *Ac* locus that can be explained on the basis of a chromosomal breakage mechanism, lead one to suspect that some kind of chromosomal breakage and fusion mechanism may likewise be responsible for these reverse mutations.

Because c^{m-1} and c^{m-2} are very different

in their phenotypic expressions, however similar they are in other respects, it should be stressed that visible mutations at the c^{m-1} locus always result in the expression of a full dominant phenotype. Varying expressions of color intensity to either side of that expected from a single dose of normal *C* do not appear.

Mutable c^{m-2} . The second mutable *c* locus, c^{m-2} , arose in a somatic cell of a plant of the constitution *I Sh ds/C Sh ds, Ac Ac*. The plant was sectorial for this mutable *c* locus. With respect to dosage of *Ac*, the production of sections, etc., the c^{m-2} locus responds exactly as do the *Ds* and c^{m-1} loci. The phenotypic expression of mutations at the c^{m-1} and at the c^{m-2} locus is distinctly different. The mutations of c^{m-2} give rise to sectors showing great variation in color intensity, from a very faint to an intensely deep color—often much deeper than that produced by a single, double, or even triple dose of *C*. Any one intensity may appear as a mutant sector in any part of the aleurone layer on any one kernel; also, any one kernel may show a number of different mutations each having its own particular color intensity.

The very great differences in phenotypic expression of the two independently arising mutable *c* loci suggest that the normal *C* locus may be composed of at least two blocks, each having its own particular organization and function but both necessary for producing the *C* phenotype. They are not strictly complementary, for combinations of c^{m-1} and c^{m-2} in the same endosperm do not result in the production of *C* color. The mutable c^{m-1} locus is assumed to have arisen from a normal *C* locus after alterations had occurred in only one block. On the other hand, the mutable c^{m-2} arose from a normal *C* locus after alterations that involved only the second block. The c^{m-1} locus undergoes muta-

tional changes only in the one affected block, whereas the mutations of c^{m-2} involve changes only in the second block. A possible alternative to this interpretation will be considered in the concluding paragraphs of this report.

THE MUTABLE wx LOCI

Two mutable wx loci are now under investigation. Each was found as a single aberrant kernel on an ear resulting from the cross of a plant homozygous for wx ds and ac by a plant that was homozygous for Wx and also for Ac . A different male parent was involved in each cross. The first case found, wx^{m-1} , was recognized because the aberrant kernel showed a wx phenotype with Wx sectors and spots instead of the expected full Wx phenotype. Because other well marked genetic characters were involved in this cross, it could be determined that no contamination had occurred to give rise to this kernel and that the mutable wx condition had arisen at the Wx locus in the chromosome 9 contributed by the male parent. No known mutable wx loci were present in any of the stocks at the time of discovery of the original kernels of either wx^{m-1} or wx^{m-2} .

The second case, wx^{m-2} , was first observed as an aberrant kernel because it showed a recessive wx phenotype, instead of the expected Wx . No mutability was recognized in the original kernel. The mutability of the locus was recognized later, when the pollen of the plant arising from this kernel was examined.

A third case of a mutable wx locus arising from a normal dominant Wx locus was observed in the cross of a female Wx Wx , Ac Ac plant by a male wx wx , ac ac plant. The kernel was almost completely wx , with only small specks of Wx . The mutations to Wx occurred late in the development of the endosperm. This type

of mutation pattern suggested that the mutability was Ac -controlled; for two doses of Ac were present in the endosperm, and two such doses are known to give this pattern with all Ac -controlled mutable loci. The embryo in the kernel did not germinate, and so this case of an independently arising mutable wx locus has been lost.

The two recovered mutable wx loci have received only preliminary study because of their recent origin. The wx^{m-1} locus is Ac -controlled and responds to Ac in all its various aspects as do the Ds , c^{m-1} , and c^{m-2} loci. It is not known whether wx^{m-2} is controlled by Ac .

All Ac -controlled mutable loci show mutations only late in the development of the sporophytic tissues. In the sporogenous cells, such mutations may be delayed until the premeiotic or meiotic nuclei are formed. The starch in the pollen of plants homozygous for a normal wx locus stains a clear red-brown color with dilute solutions of iodine and potassium iodide. The starch grains in the pollen of plants homozygous for a normal Wx locus stain a dark blue color with this same solution. In heterozygous plants, as expected, two types of pollen grains are present in equal proportions, those staining red-brown (the wx -carrying grains) and those staining deep blue (the Wx -carrying grains). In the two original plants carrying these mutable wx loci, a normal nonmutating wx locus was present in one chromosome 9 and the mutable wx locus in the homologous chromosome 9. Examination of the pollen of these two plants gave the first indication that the primary mechanism responsible for mutations of the wx loci could be associated, at least in some cases, with chromosome breakage and fusion. In both plants, most of the pollen grains, when stained with iodine, were visually wx (red-brown) in phenotype.

In the plant carrying wx^{m-1} , however, about 2 to 3 per cent of the grains were clearly different in staining reaction. These stained blue with iodine, the intensities of color ranging from light gray-blue to the very deep blue characteristic of pollen grains carrying a normal Wx locus. A high percentage of these blue-staining grains, often more than 50 per cent, were small and only partially filled with starch granules. This contrasted greatly with the proportion of such defective grains (only 2 to 3 per cent) in the wx -staining class. The pollen of the plant carrying wx^{m-2} differed from that of the plant carrying wx^{m-1} in that the recognizable blue-staining grains were confined mainly to the defective class. Less than 1 per cent of the pollen grains in this plant were defective, and in approximately half of these the starch granules stained blue. Here, also, the intensities of color ranged from a light gray-blue to a very deep blue. Only a very few of the normal-appearing pollen grains showed any trace of blue color, and it was always very faint. This contrasted greatly with the pollen of the wx^{m-1} plant, where many of the deeply blue-staining grains were normal in appearance. The questions arise: (1) Why is the proportion of defective grains so high in the mutated class in both plants? (2) What is the reason for the various intensities of the staining reaction? (3) Why are the normally developed grains with mutated loci so different in staining reaction in these two plants?

An interpretation, subject to further testing, has been formulated. It assumes that the normal Wx locus is composed of a number of identical, reduplicated units, each unit contributing its share in the ultimate conversion of precursor starch to the amylose type characteristically produced by the Wx locus. The more units are present at the locus, the greater is the

conversion and the stronger the iodine staining reaction. Published quantitative chemical studies (Sprague, Brimhall, and Hixon, *Jour. Amer. Soc. Agric.*, vol. 35, pp. 817-822, 1943; Brimhall, Sprague, and Sass, *Jour. Amer. Soc. Agric.*, vol. 37, pp. 937-944, 1945) of the nature of the starch in endosperms having various genic constitutions of wx , Wx , and a low allele of Wx known as wx^a have given a sound basis for assuming a quantitative reaction of the Wx locus. The starch in $wx\ wx\ wx$ endosperm is composed of amylopectin. With one dose of Wx ($Wx\ wx\ wx$ constitution), about 18 per cent of the starch is synthesized to amylose, the remainder being amylopectin. With two and three doses of Wx , more starch is synthesized to amylose, but not in proportion to dosage (20 and 22 per cent, respectively). Tests of wx^a , however, have shown a proportionality with dosage. Here, one dose of wx^a ($wx^a\ wx\ wx$ constitution) gives 0.65 per cent amylose, and two and three doses give 1.20 and 2.40 per cent, respectively. These published results suggest that the number of reduplicated units in the normal Wx locus may be high enough to approach, in a single dose, an effective utilization of the available substrate to be converted. This is apparently not so for a single dose of the wx^a allele. Here, too few units may be present and much of the available substrate may be unutilized. This available substrate could be utilized if more gene units were present. Added doses of the wx^a allele could accomplish this. On the assumption that different numbers of gene units are present in the Wx and wx^a alleles, it is possible to explain the different dosage responses of these two alleles.

According to the hypothesis that the normal Wx locus is composed of a number of reduplicated units, it is assumed that the mutable wx loci arose from a

normal *Wx* locus by loss of units from the locus. The remaining units are too few to produce a visual change in the color of the starch when stained with iodine. If the mutation process results in an increase of units within the locus, enough amylose may be produced to give a visible color reaction with iodine. Since the *wxa* locus gives a light orchid stain in the endosperm and a lighter stain than full *Wx* in the pollen grain, the number of residual units in the two mutable *wx* loci should be even lower than that present in *wxa*. Because individual mutations occurring at the *wx^{m-1}* and *wx^{m-2}* loci are not alike, but result in each case in the production of one particular intensity of color reaction, it must be assumed that the increase in the number of units at any one mutation is not constant; some mutations result in only small increases, others in larger increases in number of units, and so on. The various grades of intensity of staining reflect these variable increases in the numbers of units: the more units are present, the more amylose is produced, and consequently the more intense is the staining reaction.

It is believed that the defective grains in the mutated class could arise as the consequence of a breakage-fusion mechanism that results in U-shaped dicentric chromatids and an acentric U-shaped fragment, as described for the *Ds* mutations. If this break occurred within the *wx* locus or to the left of it, the dicentric chromatid could have more amylose-producing units in it than either one of the original chromatids: increased or doubled in the first case, and doubled in the second case. Breakage of this bridge in the succeeding anaphase could occur nonmedially and give rise to one broken chromatid having all these increased numbers of units and a sister chromatid having none. Suc-

cessive breakage-fusion-bridge cycles could double, triple, quadruple, etc., the number of units. Only a few mitoses would be required for building up large increments of such units. Are the defective *Wx*-staining grains the result of this mechanism? Are they defective because they have lost segments of the short arm of chromosome 9 that always accompany the formation of such dicentric chromatids? Will a deficiency of one-half to two-thirds of the short arm of chromosome 9 give rise to a detectable pollen grain, and will it be of this particular defective type?

An affirmative answer to this last question has been obtained by examining the pollen of *Wx Ds/Wx Ds*, *Ac Ac* plants. This pollen should all be *Wx*-staining except for the grains produced after a premeiotic or meiotic *Ds* mutation; these should be deficient for at least two-thirds of the short arm of chromosome 9. If, in these grains, some starch granules are formed, the starch should give the *wx* staining reaction, because a *Ds* mutation would place the *Wx* locus in the acentric fragment. This fragment is usually lost to the nucleus in the division following the *Ds* mutation. The dicentric chromosomes in the nuclei formed after a *Ds* mutation should have no *Wx* locus at all. It has previously been determined, through other types of experiments, that a deficiency of the *Wx* locus will give rise to starch of the *wx* staining type. By examination of the pollen of *Wx Ds/Wx Ds*, *Ac Ac* or *Ac ac* plants, it has now been determined that such small, partially filled, *wx*-staining grains are present in the relatively high numbers expected. Only rarely does one find such grains in the pollen of *Wx Wx ac ac* plants, and those found may arise from occasional adjacent fusions of broken ends after crossover breakage at meiosis. There is no obstacle,

then, to considering that the defective Wx -staining grains in the pollen of the wx^{m-1} and wx^{m-2} plants have arisen as the consequence of dicentric chromatid formation followed by the breakage-fusion-bridge cycle. The Wx locus is to the left of Ds , and the segment of chromosome 9 absent from these grains is even smaller than that found after a Ds mutation. The various intensities of the Wx staining reaction in the defective class of pollen grains may merely represent the various dosages of Wx units present in these grains; for various dosages could be anticipated.

The breakage-fusion-bridge cycle alone cannot explain many of the mutations from wx to or toward a Wx phenotype. Normal, functional Wx -staining pollen grains are present in the wx^{m-1} plant. It is possible that these are the reciprocal products of a breakage mechanism that sometimes gives rise to dicentric chromatids. A crisscross type of fusion may occur between broken ends after breakage at one locus in two sister chromatids. If the breakage occurs within the compound locus at unequal positions in the two chromatids, one resulting chromatid might gain enough units at a single breakage and fusion to produce a quantity of amylose starch that would give a detectable staining reaction. Several successive mutations could step up the unit dosage within the locus considerably. A graded series of staining intensities could be expected to follow such events. Examination of the mutation process in the endosperm tissues carrying c^{m-2} and wx^{m-1} suggests that such successive mutations may occur.

Examination of the kernels resulting from crosses involving the wx^{m-1} locus has been illuminating. It has shown: (1) that mutations of wx^{m-1} occur, as expected, in the endosperm tissues; (2) that these mutations differ in phenotype, some giving rise to sectors

showing only very faintly staining starch, others to sectors showing intensely blue-staining starch, with all others falling between these extremes; (3) that at least five grades of intensity can be recognized among the mutations within a single kernel; and (4) that twin sectors frequently occur, one sector showing a deeper blue stain than the sister sector. Examination of these kernels also showed that a single mutation may be followed in successive divisions by further mutations, and that these latter result in starch with either an increased or a decreased staining intensity. In other words, the mutation process may result in changes in either direction: toward full dominant or toward full recessive. A few sectors arising as the consequence of the breakage-fusion-bridge cycle were present in these kernels. Large areas within these sectors gave the wx staining reaction, but there were subsectors showing the Wx staining reaction. Within these subsectors, a wide variation in intensity of staining reaction was observed. The spatial relations of the cells showing these varied intensities of staining reaction were instructive; for adjacent areas probably arising from sister cells and showing inverse relations of color intensities were most obvious. The differences in intensities of staining reaction in these twin areas ranged from slightly detectable to pronounced, with extremes showing a deep blue adjacent to a red-brown (wx). This latter observation strongly supports the interpretation of the relation of dosage to the intensity of the staining reaction. Dosage changes appear to be responsible for the changes in staining reaction within these sectors having dicentric chromatids; the greater the dose, the more intense the staining reaction, and vice versa. It is realized that increments or decrements of units must fall within the range that can

give visually distinguishable changes in intensities of color.

It is not mere speculation, therefore, to consider that the quantitative grades of mutation occurring at the mutable wx loci may follow increases in the number of identical units within a depleted locus that is normally composed of a relatively large number of such reduplicated units. The question arises, What is the primary mechanism responsible for such increases and decreases, and do all mutable loci reflect this same general mechanism?

CONCLUSIONS

It may be premature to consider in detail the question asked in the concluding sentence above. Because so many mutable loci have recently appeared in the maize cultures, because in many respects they all behave in very much the same way, and because this behavior is similar also to that described for other mutable loci both in maize and in other organisms, it may be profitable to review briefly the pertinent facts about the cases described in this report, in order to ascertain the similarities and dissimilarities among these cases. They involve the mutable locus Ac and the mutable loci it controls— Ds , c^{m-1} , c^{m-2} , and wx^{m-1} .

Ds stands alone in that the chromosomal consequences of a mutation at this locus are known. The detectable Ds mutations unquestionably arise as a consequence of some mechanism that either brings about breakage and fusion between sister chromatids at the Ds locus or simulates this mechanism in its consequences; for dicentric chromatids are produced after a Ds mutation. In the pattern of mutations, c^{m-1} and Ds are strictly comparable. Variegation patterns produced by Ds mutations in $C ds/C ds/I Ds$, $Ac ac ac$ constitutions can be so similar to variegation patterns

produced by c to C mutations in $c^s c^s c^{m-1}$, $Ac ac ac$ constitutions that they may be indistinguishable by mere observation of the kernels. Yet the formation of dicentric chromatids alone cannot explain most of the observed c^{m-1} mutations. This likewise applies to the c^{m-2} mutations and to many of the mutations of wx^{m-1} . Some form of chromosome breakage and fusion, however, may be involved.

As stated earlier, mutations of c^{m-1} and c^{m-2} differ greatly in phenotypic expression. Visible mutations of c^{m-1} give rise each time to the full dominant expression expected from a single dose of C , whereas those of c^{m-2} are quantitatively expressed, with color intensities varying from faint to extremely deep. The normal C locus is known to give dosage effects: the more whole C loci present, the greater the depth of color. By means of duplications of the short arm of chromosome 9, it has been possible to observe effects of doses up to and including six C loci. With the highest dose, the color of the aleurone is unusually deep. Some of the mutations of c^{m-2} result in intensities greater than that shown with three doses of the normal C locus, whereas others are so faint that they obviously have produced much less pigment than is produced by a single normal C locus. The various consequences of mutations of the c^{m-2} locus are in complete agreement with the hypothesis that the mutations result from graded increases in the number of units within a depleted locus and that they express themselves phenotypically by graded increases in the substance or substances responsible for the phenotypic character. The observed mutations of c^{m-1} do not lead to this hypothesis, for they show no quantitative subdivisions. Similarity of the c^{m-1} mutations to Ds mutations in basic response to Ac , and their conformity to the general pattern of the Ac -controlled

mutations that do give quantitative effects, has led to formulation of a subsidiary hypothesis rather than abandonment of the general hypothesis. The validity of this subsidiary hypothesis is subject to tests that are now being conducted. Assuming the fundamental mutation process to be similar for all *Ac*-controlled mutable loci, the genes in the mutating c^{m-1} block of the *C* locus could be related to some chemical process that requires a threshold number of units for expression to be fulfilled; or it may be that the mechanism bringing about a change in units at this mutable *c* locus assures a specific increase in number of units.

The mutations occurring at the c^{m-2} and wx^{m-1} loci are amazingly similar in every respect. The mutations fall into a graded quantitative series in both cases. Also, both give twin or adjacent sectors showing the same grades of contrasting intensities of expression of the dominant character. There can be little doubt that the same mechanism is responsible for the mutations occurring at these two different loci. With regard to the wx^{m-1} locus, there is evidence for believing that the unit number within the locus may be responsible for the expression of unit increases of the dominant phenotypic expression. Considering the obvious similarities between the two cases, it would be difficult to avoid concluding that the same conditions apply to mutations at the c^{m-2} locus. In this connection it may be recalled that mutations at the *Ac* locus likewise suggest a mutation mechanism involving changes in the numbers of units at the locus.

Because many of the mutations occurring at the two mutable *c* loci, the wx^{m-1} locus, and the mutable *Ac* locus do not result in dicentric chromatids, as do *Ds* mutations, and do not lead to detectable gross chromosomal aberrations, the mechanism, if it is a breakage phenomenon,

must restore the normal chromosome morphology. Unequal breakage within the locus, followed by the crisscross type of fusion mechanism, could accomplish this end.

Any mechanism that gives rise to an increase in numbers of units at a locus might also give rise to the reverse condition; that is, to chromatids with decreased numbers of units. If so, chromosomes with loci having various unit numbers should appear as isolates in these cultures. Some of these isolates should show more visible mutations than others under given conditions. The frequency of visible mutations would depend upon the initial number of units present in the locus before mutation occurred. The more units were present, the greater would be the chance that the increase in the units during any one mutation would be sufficient to produce a visible effect; and the converse would also be true. Isolates from the different mutable loci, showing just these expected variations in the rates of visible mutations, have been obtained. The term "state of the locus" applies as well to the mutable loci controlled by *Ac* as to the *Ac* locus itself. What has been termed a high-state locus gives high rates of mutation, and a low-state locus gives low rates of mutation. It is possible, therefore, that the number of units present at a mutable locus is correlated with the state of the locus as well as with the expression of visible effects. It is a matter of degree. If the initial number is high, but not high enough to produce a visible effect, the state of the locus may be considered high. Conversely, if only a few units remain in the locus, the state of the locus may be considered low.

The above conclusions are supported by the many observations that have been made especially of the chlorophyll-producing types of mutable loci. As mentioned in

previous reports, the motivation for this study of mutable loci was the observation of twin sectors, apparently arising from sister nuclei, that showed inverse rates of visible mutations. The most extreme of these twin sectors showed a mutation to dominant in one sector, and in the sister sector either a complete recessive or a very much reduced rate of mutation as compared with that in the surrounding tissue. In these observed cases of twin sectoring, it is obvious that the factor or factors controlling the rate of mutation and the visible mutations themselves are of the same general nature, if not actually different resultants of the same mechanism. The factor responsible for twin sectoring acts at a mitosis, and the apparent result is that one chromatid gains something that the sister chromatid loses. In the interpretation given, this gain and loss are considered to be an increase and a decrease, respectively, of identical units at the locus, which, in turn, controls not only the appearance of visible mutations but also the state of the locus as reflected in the time and frequency of occurrence of future visible mutations.

The above interpretations are being used as a working hypothesis in continuing studies of mutable loci. The evidence at present points toward the presence of reduplicated units within a locus, these units often expressing themselves in a quantitative manner through unitary action on substrates responsible for phenotypic characters. The hypothesis that the

phenotypic expression of a locus depends upon the number of such units present in any one chromosome, and that some mechanism or mechanisms can alter this number and give rise to visible mutations, is both sufficiently simple and sufficiently integrative to afford precise direction in some types of experimentation. It is believed that this approach to one phase of the over-all mutation problem may be productive, even though it is realized that the details will need clarification and may be subject to degrees of modification. With so many mutable loci behaving in very much the same manner, it is unlikely that many different, unrelated mechanisms are involved. It is more likely that one general type of condition exists in all these mutable loci and that this condition may be altered in any of these loci by one kind of mechanism.

In this report, it has not been possible to include a discussion of the many other observations and conclusions that are relevant to the subject, such as the reestablishment of a mutable locus, the behavior of *Ac*-controlled mutable loci when two or more are present in the same nucleus, or the changes at the *Ac* locus that often appear to accompany an *Ac*-induced mutation. Nor has it been possible to describe the accumulated evidence on some of the non-*Ac*-controlled mutable loci, or to mention the many new mutations that are constantly arising in these cultures. These studies are being continued and will be reported later.

THE GENE

M. DEMEREC, B. WALLACE, AND E. M. WITKIN

Our group worked primarily on problems dealing with analysis of the mutagenic properties of various chemicals. The studies on *Drosophila* were continued with carcinogens and compounds related

to carcinogens, while the group working with bacteria made investigations designed to improve the methods used in detecting the mutagenic properties of chemicals, and completed such tests with a number of

chemicals. Other bacterial experiments dealt with population dynamics in cultures of two strains grown together.

We were assisted in this work by Misses M. N. Crippen, J. Flint, B. Hawxhurst, E. Lively, S. L. Rohrer, and H. Spring, Mrs. J. S. Buchanan, and Mr. P. A. Peterson. The work was aided by a grant from the American Cancer Society, recommended by the Committee on Growth of the National Research Council. Dr. Witkin participated in the research as a fellow of the American Cancer Society.

MUTAGENIC POTENCY OF CARCINOGENS AND CHEMICALLY RELATED COMPOUNDS

Additional evidence has been obtained by M. Demerec and B. Wallace concerning the mutagenic activity of the chemicals discussed in last year's report (Year Book No. 46 [1946-1947], pp. 127-131). The number of tests for mutagenic effect has been increased, and new observations have been made on induced breaks in chromosomes.

It is well known that chromosomal aberrations so seldom occur spontaneously in *Drosophila* that the appearance of any in a treated series would in itself strongly indicate genetic effectiveness of the treatment. For this reason observations on chromosomal aberrations are important in the determination of mutagenicity in chemicals, particularly when the effectiveness of the chemicals is low. So far our tests for chromosomal aberrations have been limited mainly to the material from experiments on mutations. It is known that chromosome rearrangements frequently affect genes located in proximity to breaks, and give rise to either lethal or visible mutations. Therefore if a chemical used in experiments induces both lethals and chromosome breaks, it is to be expected that they will frequently be found

together. Consequently, chromosomal aberrations should be more frequent among lethals than among nonlethals.

All lethal mutations obtained in our experiments were tested for chromosomal aberrations by genetical methods; and if the results were positive, they were checked by analysis of salivary-gland chromosomes. Aberrations were found in material treated with (a) 1,2,5,6-dibenzanthracene (3 among 43 tested), (b) 3,4-benzpyrene (1/24), (c) 1,2-benzanthracene (1/8), (d) *p*-hydroxyazobenzene (3/44), (e) *p*-dimethylaminoazobenzene (1/12), and (f) 2-amino-5-azotoluene (1/6). All but (c) are carcinogens; and all but (e) and (f) produced a significant increase in mutations.

Sixteen hydrocarbons and azo compounds, in all, were tested in this series of experiments—7 that are known to be carcinogens, and 9 in which carcinogenic activity has not been detected. Of the 7 carcinogens, 6 were found to be mutagenic for *Drosophila*, and 1 not; whereas of the 9 noncarcinogens, 2 were mutagenic, 6 were not, and 1 is still doubtful. It is evident that the carcinogenic and mutagenic properties coincide to a considerable degree.

During our experimentation it was observed that different males treated at the same time quite frequently showed different results; that is, genetic changes could be induced in some males more readily than in others. It was noticed also that similar treatment in different experiments occasionally brought about different results. Investigations have suggested that these differences may be due to differences in genetic constitution.

INDUCTION OF MUTATIONS IN BACTERIA BY CHEMICALS

Work on the induction of mutations in *Escherichia coli* by chemical agents was

continued by E. M. Witkin. The principal objectives of this phase of the investigations were: (1) extension of preliminary screening tests to characterize various compounds with regard to mutagenic activity; (2) development of more efficient techniques for preliminary screening; (3) investigation of "delayed" mutations induced by chemicals; and (4) development of several independent methods for demonstrating the activity of mutagenic chemicals.

1. *Preliminary screening tests.* The "zero-point mutation" method, previously described (see Year Book No. 46), has been used to explore the mutagenic potency of various biologically active chemicals. The method involves suspension of resting bacteria of strain B/r in toxic concentrations of the chemical, and subsequent determination of the proportion among the survivors of mutants resistant to bacteriophage T₁, as compared with the proportion of resistant mutants in an untreated sample of the same culture. Controls were developed to determine whether phage-resistant and phage-sensitive bacteria differed in sensitivity to the toxic action of the chemicals, since differential rates of killing might simulate the results to be expected if zero-point mutations were induced. Similarly misleading results might be obtained if the sensitive bacteria, after treatment with a chemical, were considerably delayed in their rate of infection by the phage; and so experiments were made to measure the rate of infection of treated bacteria by T₁. The conclusion that mutations had been induced by a given chemical was felt to be justified only when these and other possible complicating factors could be eliminated. The screening tests to date have indicated that the following chemicals are capable of inducing zero-point mutations: sodium desoxycholate, acriflavine, pyronin (all re-

ported in Year Book No. 46), caffeine, and colchicine. Mutagenic activity has been suggested, but not yet conclusively demonstrated, for a number of other compounds, including sodium chloride. Negative results have been obtained with methyl green, basic fuchsin, and a number of nontoxic compounds including methylcholanthrene endosuccinate.

2. *Development of a more efficient screening technique.* The zero-point-mutation method, which has until recently been used as a means of screening compounds for mutagenic activity, suffers from a number of disadvantages. The most important of these is the rather restricted range of survival of the chemically treated bacteria over which the technique can be used. Ordinarily, the survival rate of a treated culture must be no greater than 1 per cent (since high survival is usually accompanied by weak mutagenic action) and no less than about 0.05 per cent (since the absolute number of survivors must be large enough to permit the detection of induced mutants). The survival level, under standard conditions of treatment with most toxic compounds, has been found to be extremely variable and unpredictable. Thus, since it is necessary to perform a number of full zero-point experiments before hitting upon one that falls within the practicable survival range, the method has proved rather inefficient. Other disadvantages of the zero-point method are the relatively elaborate controls required to eliminate such complicating factors as differential survival and altered rates of infection by phage.

Very recently, experiments were begun to explore the possibilities of another method of screening compounds for mutagenic activity. This method is based on analysis of the distribution of mutants in a series of liquid cultures inoculated with survivors of exposure to toxic chemicals.

It is well known that, in a series of liquid cultures inoculated with untreated bacteria, the number of mutants from culture to culture, after growth is completed, is subject to wide fluctuation. This is due to the clonal grouping of spontaneous mutants, whereby a mutation arising exceptionally early in the growth of a culture will reflect itself in a final crop of mutants very much higher than the average. The number and magnitude of these "jackpot" cultures should be increased significantly, owing to either zero-point or delayed mutations, or both, in a series of cultures inoculated with survivors of exposure to a mutagenic agent. This method has been validated by Newcombe (in press) for ultraviolet radiation; and preliminary results with X-rays in our laboratory indicate that it is extremely efficient. This technique is practicable over a far greater range of survival than the zero-point method, and is relatively independent of possible complicating factors. We plan to use this technique as a qualitative screening test in place of the zero-point method.

3. *Delayed mutations.* Demerec (see Year Book No. 44 [1944-1945]) and Demerec and Latarjet (see Year Book No. 45 [1945-1946]) have shown that mutations to resistance to bacteriophage induced in *E. coli* by radiation are of two types: "zero-point" mutations, which are induced and expressed phenotypically in resting cells; and "delayed" mutations, which are expressed only after the survivors of the exposure to radiation have been permitted to divide. When high doses are used, these delayed mutations continue to be expressed for many cell generations after the treatment. Zero-point mutations are known to occur under the influence of mutagenic chemicals. One of our objectives has been to determine whether, and to what extent, delayed

mutations also are induced by exposure to chemical mutagens.

The techniques used to detect delayed mutations to phage resistance were essentially those described by Demerec in connection with radiation-induced mutations (see Year Book No. 44). The bacteria were exposed to the chemical as usual, then carefully washed, and a large number of identical samples spread upon nutrient-agar plates. At various time intervals, depending on the previously determined growth rate of the treated bacteria under these conditions, random sets of these plates were removed from the incubator, and used to determine (a) the numbers of viable cells on the plates, and (b) the numbers of clones of T1-resistant mutants on the plates, at the time of test. To determine the numbers of mutant clones, the plates were sprayed with a fine aerosol mist of bacteriophage, so that the bacteria were not agitated or moved, and then incubated. Under these conditions, each clone of phage-resistant bacteria, regardless of its size, will give rise to a single colony. This permits a direct reading of the rate of appearance of new mutant clones during the division on the plates, and thus a direct determination of mutation rate. For sodium desoxycholate-treated bacteria, the rate of appearance of T1-resistant mutants in the first division following treatment was found to be about 75 per 10^8 new bacteria produced; the rate for untreated bacteria during the corresponding division is about 12 per 10^8 new cells produced. Delayed mutation appeared, therefore, in the first division following exposure to sodium desoxycholate. In the second division following treatment with this compound, however, the rate of appearance of new mutant clones did not differ significantly from the rate in the corresponding division of untreated bacteria. Thus the delayed effect does not

seem to persist over a large number of generations, as it does in the case of high doses of radiation. Other compounds found to show mutagenic activity on the basis of screening tests will be investigated with regard to the production of delayed mutations.

4. *Independent methods of demonstrating mutagenic activity.* Since the beginning of these investigations, resistance to bacteriophage T1 has served as the sole genetic index of the mutagenic activity of chemicals. Availability of at least one other independent mutational class would permit confirmation of results obtained using the phage system, and also measurement of the genetic specificity of the effects observed. The "reverse" mutations from biochemically deficient strains to the prototrophic habit seemed, a priori, to constitute a mutational class which would, like phage resistance, be amenable to quantitative investigation. Consequently, preliminary work was done to explore the spontaneous reversions of biochemically deficient strains of *E. coli*.

Several strains were obtained from Doermann, and many others were isolated from strain B/r by a modification of the Lederberg layering technique. When cultures of these deficient strains were grown in complete medium, carefully washed, and plated out in large samples (10^7 or 10^8 cells) on minimal agar, a small number of distinct colonies usually appeared after incubation. Cultures established from these colonies were capable of full growth in minimal medium. The reverse-mutation rates appeared to vary over a considerable range for different strains (10^{-6} to 10^{-9} mutations per bacterium per generation).

In the course of this preliminary work it became clear that the reverse-mutation behavior of most of these strains was not so clear-cut as might appear. Microscopic

observation and other techniques showed that, in spite of the inability of these strains to grow to turbidity in liquid minimal medium, a considerable number of divisions usually occurred on minimal agar. The number of divisions, moreover, varied with the inoculum size in a rather complicated way. These factors can lead to very large distortions in estimating mutation rate, and often operate in a direction that tends to simulate mutagenic effects. The residual division on minimal-agar plates, although it represents a serious obstacle, can probably be controlled. If it is due to growth-factor traces stored within the bacterial cells, a preliminary incubation in liquid minimal medium before plating out should lead to exhaustion of the available traces. If it is due to contamination of the agar by growth-factor traces, more thorough methods of washing the agar will solve the problem. Once these difficulties are overcome, it should be possible to utilize reverse mutations from biochemically deficient strains as an additional index of mutagenic activity of chemicals. These strains should also prove useful because the availability of numerous deficient strains having widely different reverse-mutation rates makes possible an analysis of the relation between spontaneous mutation rate and susceptibility to induced mutation.

POPULATION DYNAMICS IN BACTERIA

The population dynamics of mixed cultures of strains B and B/r of *Escherichia coli* (see Year Book No. 45) were further investigated by Witkin. Strain B/r, a radiation-resistant variant isolated as a spontaneous mutant of strain B, exhibits essentially the same growth behavior as the parent strain during the phases of growth represented in the 24-hour cycle. Thus, in repeated daily sub-

cultures, artificial mixtures of the two strains maintain the proportions initially introduced. If such mixtures are permitted to age without subculture, however, a marked population shift occurs, apparently owing to intense selection against the B/r component. Incubation at 37° C. for a period of 2 to 3 weeks is often sufficient to eliminate the B/r component entirely. An investigation of the basis of this selection against strain B/r, and of the conditions under which it operates, may be useful for several reasons: (1) an important physiological difference between strains B and B/r is indicated, which may be related to the mechanism of radiation resistance; (2) the application of certain theoretical concepts of population dynamics may yield genetic information not obtainable by other methods; (3) the possibility of controlling selection factors in genetically mixed populations of bacteria may be of practical importance—for example, in clinical and epidemiological applications.

It seemed desirable, first, to determine whether the drop in proportion of B/r cells in aging mixtures with strain B was due (a) to selective death of B/r, (b) to selective division of B, or (c) to a combination of the two. An analysis was made of the pattern of division and death in aging pure cultures of B and B/r, and in aging mixtures of the two strains.

It was determined first that strains B and B/r, and mutants of these strains resistant to bacteriophage T₁ (B/1 and B/r/1), behave identically with regard to selection in aging mixtures. Thus, mixtures of B and B/r, of B/1 and B/r, or of B and B/r/1 all gave the same rates of population shift when maintained under similar conditions. Instead of using B and B/r in these experiments, therefore, the mixtures were made up with either B and B/r/1 or B/1 and B/r, so that one com-

ponent of the mixture was marked by resistance to bacteriophage T₁. In determining the proportions of the two components of the mixtures, it was possible to use phage T₁ as a simple means of separating the components, instead of the more cumbersome and less sensitive double-irradiation treatment previously used. The use of phage resistance as a genetic marker has numerous other advantages, such as permitting the separation of those radiation-resistant organisms initially introduced into the mixture from those arising spontaneously by mutation from the B component during the course of the experiment.

A series of nonaerated 30-ml. broth cultures, each inoculated with about 100 bacteria, was set up and incubated for 24 hours at 37° C. There were 10 cultures each of strains B, B/1, B/r, and B/r/1. After 24 hours, 0.03 ml. of one of the B cultures was added to each of 5 of the B/r/1 cultures, and the same amount of one of the B/1 cultures was added to each of 5 of the tubes containing B/r. Thus two sets of mixtures were set up, each having at the start 99.9 per cent B/r or B/r/1 to 0.1 per cent B/1 or B. The pure cultures and the mixtures were maintained at 37° C., in closed moist chambers to prevent evaporation, for a period of 5 weeks. Starting when the cultures were 1 day old, and every 2 days thereafter, the following tests were made, using for each determination two samples of each type of pure culture and each type of mixture.

1. Viable counts were made by dilution, plating, and counting of colonies. These counts gave the number of living bacteria per milliliter of culture (the criterion of life being, throughout these experiments, the ability to give rise to a visible colony on nutrient agar). In the mixtures, viable counts were made for each component. In a mixture of B and B/r/1, for example,

one set of assays was performed on ordinary nutrient-agar plates, giving the viable count for both types of bacteria, and another set on Ti-coated plates, on which only the B/r/1 component could grow. Thus the absolute viable count for each component, as well as the relative proportions, could be calculated.

2. For each of the pure cultures and mixtures, a total count was made. The total count represented the number of bacteria, both living and dead, present in the culture at the time of test, and was determined both by turbidometric measurement, using Doermann's photoelectric nephelometer, and by microscopic count, using a Petroff-Hauser bacterial counting chamber under dark-field illumination. Since results obtained by the two methods agreed well, the nephelometric readings were used exclusively in the latter half of the experiments. Autolysis is rare in *E. coli*, and so an increase in total count between two times of determination is a good measure of the amount of division that has occurred in the interval, whereas the difference between total and viable counts represents the number of dead cells.

Curves for the total and viable counts of pure cultures and mixtures over a 5-week period were established, giving a clear picture of the dynamics of death and division in these cultures. To summarize these results: (1) Viable counts for all the pure cultures and mixtures remain relatively stable, showing no significant differences among themselves, and fluctuating about a mean of approximately 10^8 per milliliter. (2) Total counts for pure cultures of strains B and B/1, on the one hand, and B/r and B/r/1 on the other, show significant differences in the extent of division in aging cultures. Strains B and B/1 divide at a constant logarithmic rate for a period of about 12 days, with the result that the total count increases by

a factor of about 10 during this period. Strains B/r and B/r/1 divide at a lower rate for a shorter period of time, with the result that the total count increases by a factor of 3. In all these strains, division is almost exactly counterbalanced by death, since the viable counts remain constant.

(3) The total counts of the mixtures show a composite pattern. For the first 6 or 7 days, the curve is identical with that of strain B/r (the mixtures having been started with 99.9 per cent B/r). About the 7th day, when the B component has climbed to about 50 per cent of the viable count, the total count begins to rise at the rate characteristic of strain B, reaching the tenfold increase and then leveling off a few days later than the pure cultures of strain B. Thus, it is apparent that part of the population shift in aging mixtures is due to the more active division of strain B under these conditions. That the more dramatic features of the shift are due to selective death of strain B/r is shown by the following: (4) The viable counts of the B and B/r components of the mixtures show that the B component increases rapidly in the first few days, rising from 0.1 per cent (10^5 cells) to 99.9 per cent (10^8 cells) by the 10th day. The viable count of the B/r component decreases correspondingly in the same period. The viable count of the B/r component continues to drop steadily, well beyond the period of active division of the B component. In one case it was possible to show, by plating out an entire culture, that 5 weeks after the start not a single B/r organism remained in the culture.

Part of the selection against B/r in mixtures can be explained by the more active division of the B strain. The medium, apparently, can support a fixed number of viable cells. If division increases this number, a compensating number of

bacteria will die. In the aging mixtures, B continues to divide long after B/r stops. Thus, as the division of B raises the total above the viable maximum, death of both B and B/r will occur. The B/r deaths, however, are not counterbalanced by division. If this were the whole story, the selection against B/r should stop when the active division of the B component stops, at about 15 days. The fact that B/r continues to drop to complete extinction suggests that another mechanism, such as an active toxic product, may be at work. It is also possible that the disappearance of B/r organisms is due to the action of a transforming agent. To test these possibilities, the following experiments were made: (1) B/r cells were suspended in filtrates of B cultures of various ages; and (2) B/r and B were incubated in a dialysis system, with the two strains separated only by a cellophane membrane. In neither experiment was there any drop in the viable titer of the B/r cells. Therefore no filtrable or dialyzable toxin or transforming agent seems to be involved.

In another experiment, the problem of reverse mutation from B/r to B was approached through selection phenomena. Ordinarily such reversions would be exceedingly difficult to detect, and it had

not been possible to make any statement about their existence or frequency. A very small number of B bacteria (less than 10) was added to each of four fully grown B/r cultures, and the four tubes were incubated at 37° C. for 3 weeks. At the end of this time it was found that all four tubes contained almost 100 per cent B cells. Thus, a very small number of B organisms in a B/r culture can overgrow the B/r cells in a few weeks. If pure B/r cultures contained even a few B reversions (having all the properties of the original B), they should show instability on prolonged incubation, owing to overgrowth by B. In fact, however, B/r cultures are perfectly stable in this respect. Therefore reverse mutation from B/r to the original B probably does not occur.

Recent experiments indicate that the intensity of selection against B/r in mixture with B is extremely sensitive to conditions of aeration; and it may be possible to eliminate, or even reverse, the selection by controlling this factor. Another area under investigation is the interplay between selection against B/r and mutation from B to B/r during the period of active division. The use of phage resistance as a genetic marker greatly facilitates this type of study.

INTRACELLULAR GROWTH OF BACTERIOPHAGE

A. H. DOERMANN

The growth of bacteriophage takes place during the time between infection of the host cell and its lysis. During this "latent period" a hundred-fold or even greater increase in phage particles may take place. With certain phages the latent period may be as short as 13 minutes. Prior to these experiments no one had succeeded in liberating any phage particles before the end of the latent period. It had thus been impossible to determine directly the num-

ber of phage particles inside the cell at any one stage of the latent period.

The problem attacked in the past year was that of developing a method for liberating the viral contents of infected bacteria at will. The material used for this study was *Escherichia coli*, strain B, and several phages of the T system (Delbrück, *Biol. Rev.*, vol. 21, p. 30, 1946). That it might be possible to develop such a method with this material was suggested by informa-

tion from several sources. Delbrück in 1939 (*Jour. Gen. Physiol.*, vol. 23, p. 643) described a phenomenon which he called "lysis from without," which depended on rapid infection of a cell with a large number of phage particles. Doermann further observed (unpublished) that the addition of a large number of T6 phage particles to bacteria already infected with T4 would, under some conditions, cause lysis of the cells and liberation of T4 phage particles before the expiration of their normal latent period.

MATERIALS AND METHODS

Preliminary experiments showed that lysis from without, though it did yield new phage before the end of the latent period, was too slow a process to give a true picture of the contents of a phage-infected cell at a specific time. It was hoped that by stopping phage growth, more time could be given the lysing agent to accomplish its purpose. Low temperature could not be used to stop phage growth, because lysis from without does not proceed sufficiently well under this condition. Metabolic poisons were tried, and the methods here reported involve the use of two of them, 5-methyltryptophane (5MT) and cyanide (CN^-). (The 5MT was obtained through the courtesy of Dr. M. L. Tainter.)

In order to employ 5MT successfully, it was necessary to use a medium in which the tryptophane concentration was known and could be controlled, but which could be fortified so as to give a moderately large phage yield per infected cell. The medium finally employed contained inorganic salts, glycerol, acid-hydrolyzed casein, and tryptophane when desired. The acid-hydrolyzed casein was used to fortify the medium because it is a well balanced source of amino acids but is completely lacking in tryptophane. In addition, the medium con-

tained 10 μg . of gelatin per milliliter, in order to prevent surface inactivation of phage (Adams, *Jour. Gen. Physiol.*, vol. 31, p. 417, 1948), and 200 μg . of Tween-80 per milliliter to lower the surface tension and thereby facilitate the pipetting of small volumes. This medium will be referred to as the growth medium, or GR.

It was found that even in this medium, which was completely lacking in tryptophane, it was impossible to inhibit the B strain of *E. coli* with 5MT, presumably because this strain was able to synthesize tryptophane in amounts sufficient to overcome the inhibition. A tryptophane-deficient mutant, B/1, was therefore used. Later experiments were made with a mutant still further altered by making it resistant to radiation. The radiation-resistant strain (B/r/1, supplied by Dr. Witkin) is desirable because it has less tendency than the radiation-sensitive form to give rise to chains of bacteria in the media used here.

In addition to the GR medium described above, a lysing medium was used. This consisted of GR, with the addition of a high-titer T6 stock (final concentration, about 4×10^9 particles per milliliter) and either 5MT or CN^- . Cyanide was used at a concentration of 0.01 M, and 5MT at 100 μg . per milliliter. When 5MT was used in the lysing medium, tryptophane was omitted. Under the conditions of the experiments, these two lysing media had no effect on the titer of free phage particles.

EXPERIMENTS WITH LYSIS PROCEDURES

In the first experiments, 5MT was used to stop phage growth. B/r/1 cells in the exponential growth phase were concentrated by centrifugation to about 10^9 cells per milliliter. To these concentrated bacteria a T4r phage was added and incubated for 1 to 2 minutes with aeration.

This allowed time for at least 80 per cent of the phage to be adsorbed by the bacterial cells. Then a 40-fold or larger dilution was made in GR containing anti-T4 rabbit serum. This inactivated most of the residual free phage. After several minutes' exposure to the serum, a further large dilution was made in GR to reduce the serum concentration to one of relative inactivity. The resulting culture will be referred to as the source culture; the entire experiment was carried out with infected bacteria from the source culture. The titer of infected B/r/1 in this tube was approximately 10^5 per milliliter.

From the source culture a further dilution of 1:20 in GR was made at any time before the end of the normal latent period. The resulting culture, containing approximately 5×10^3 infected bacteria per milliliter of GR, was used for determining the end of the normal latent period, and for estimating the average yield of phage per infected bacterial cell. It will be called the control growth tube. In the experimental procedure, a number of 20-fold dilutions were made from the source culture at precise time intervals in lysing medium containing 5MT.

The titers of these various cultures were determined by plating aliquots in a thin layer of 0.7 per cent agar containing B/6, a mutant of B resistant to T6 but sensitive to T4. In this way the number of T4 particles could be estimated in the presence of a large excess of T6. Platings were made from the control growth tube before the end of the latent period and just at its end. From the former we found the number of infected bacteria, and from the latter the precise time when they began to burst. Platings were also made after the end of the latent period from aliquots of the control growth tube diluted further. From comparison of preburst and postburst

platings the average yield of phage was calculated.

Titration of the experimental cultures were made after they had been incubated in the lysing medium for 30 minutes or longer. By comparison of these titers with the preburst platings from the control growth tube, and with the average cell yield in the control culture, the fraction of the cell yield was found. This, as a preliminary working hypothesis, was considered to be the fraction of the phage present in the bacteria at the time of dilution into the lysing medium.

It should be noted that the experimental assays were not affected by the poisons. All the platings involved a dilution of the poison. Furthermore, 5MT was counteracted by the high concentration of tryptophane in the nutrient agar, and cyanide was apparently replaced by carbon dioxide quite rapidly in the plated cultures.

In figure 3 the results of typical experiments are shown by curves A and B. Curve A shows the results of applying this technique to bacteria infected by an average of 5 phage particles per cell. Curve B gives the results of single infection.

Also shown in figure 3 are the results of experiments using CN^- as the agent for stopping phage growth in the lysing medium. The experiments are comparable to those in which 5MT was employed, but with one striking difference. Both the single- and the multiple-infection curves are moved to the right by 3 to 4 minutes, indicating that at a given time less phage is liberated per cell if lysis occurs in the presence of cyanide than if it occurs in the presence of 5MT. The reason for this difference is not clearly understood. At least two hypotheses seem possible. First, perhaps the cyanide penetrates the cell faster than the 5MT. This would allow more synthesis of phage to go on with 5MT, between the time of exposure to

the poison and the time it reached the site of inhibition. In this event it would seem that using higher concentrations of 5MT would enable it to penetrate faster and cause the curve to approach the CN⁻ curve. To test this, the standard concentration was used in parallel with one 5 times as high. No difference could be detected between

phage particles, and many particles that have passed this reaction will go on to maturity before the cell breaks open. Cyanide, on the other hand, may block one of the terminal reactions in phage synthesis, and at a given time many fewer particles will have passed this reaction than will have passed the 5MT-inhibitable step.

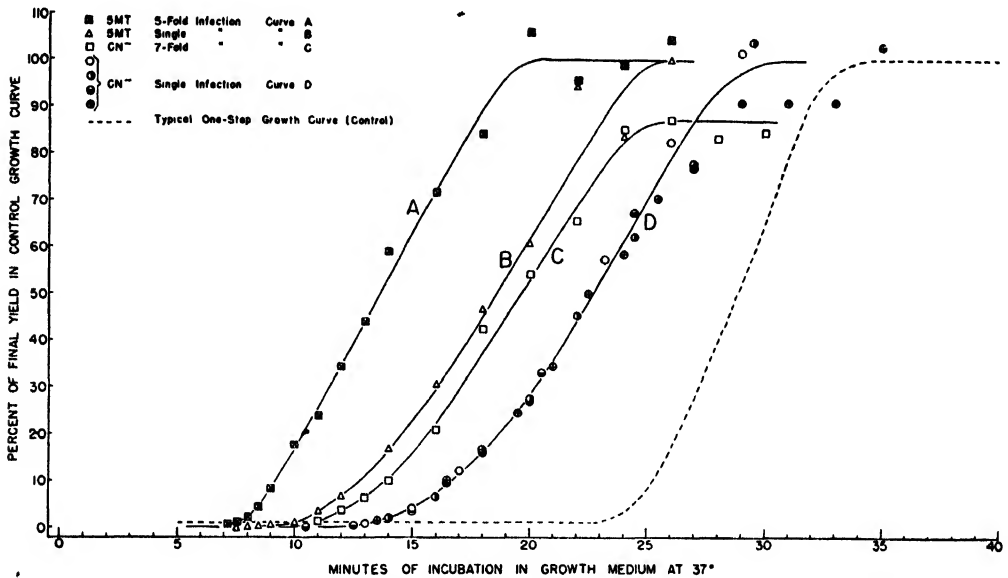


FIG. 3. Comparison of 5MT and CN⁻ as inhibitors for a lysing medium. Points for these curves obtained from platings made 30 or more minutes after dilution into the appropriate lysing medium. The abscissa of each point shows the time of incubation in growth medium that preceded this dilution.

the two curves. Moreover, 2.5 times less cyanide gave a curve identical with the CN⁻ standard-concentration curve. These experiments do not rule out the possibility that the difference between the 5MT and the CN⁻ experiments is due to a difference in rate of penetration, but they do make this hypothesis seem less probable. It also appears less likely that the reaction inhibitable by 5MT is only partially blocked by the concentration customarily used.

A second hypothesis is that the reaction blocked by 5MT is one of the earlier ones involved in the production of mature

This would explain the presence of more phage at a given time when 5MT is used than when cyanide is used.

At this point it was evident that our information was inadequate to decide whether either of these methods gives a true picture of the contents of the infected cell. Apparently an independent method was required to confirm one or the other of the above groups of data. Accordingly, experiments were made using intense sonic vibrations rather than lysis to break open the bacteria. These experiments are described in the next section.

First, however, one other result should

be mentioned. In the attempt to test whether exposure to either CN^- or 5MT caused death of the infected bacteria, a peculiar phenomenon was noticed. Cohen and Anderson (*Jour. Exper. Med.*, vol. 84, p. 525, 1946) reported exponential decrease in the number of infective centers in the presence of 5MT. In the first experiments done here to test both 5MT and cyanide in this respect, it was found that there was a very rapid increase in the number of infective centers. Further experiments showed that the increase or decrease in number of infective centers depended on the time of addition of the poison.

Both turbidimetric and plaque-count experiments support the following interpretation. If either of the poisons is added before the time when the cell contains phage particles that can be liberated in the presence of that poison, then the bacteria slowly die and do not lyse. If it is added after the time when phage can be liberated in the lysing medium containing the poison, then the cells lyse, liberating a number of phage particles. With CN^- , this number is the same as in the standard cyanide lysing medium. Thus it is only in the early stages of the latent period that it is necessary to have T6 present in the CN^- lysing medium. In the experiments described here, however, T6 was consistently used in all lysing media.

With 5MT the result is essentially the same as with cyanide, except that a few more phage particles are consistently liberated in the absence of T6 than in its presence. This result is not clearly understood, but may mean that T6 induces faster lysis than 5MT alone, and that between the times of poisoning and lysis, particles that have passed the 5MT-inhibitable stage can mature before being thrown out into the medium. It should be noted that the experiments of Cohen and Anderson are hereby confirmed and extended. In

their experiments the poison 5MT was added before the cells were infected.

EXPERIMENTS WITH SONIC VIBRATIONS

T. F. Anderson and his associates (*Science*, vol. 108, p. 18, 1948) suggested the possibility of studying the kinetics of phage reproduction by sonic disruption of cells during the latent period. Communication of Doermann's results to Dr. Anderson resulted in his proposing a joint study to determine whether sonic vibrations could be used for this purpose and to test whether the results using lysing media might be confirmed by such a study. The sonic-vibration experiments were therefore carried out in collaboration with him in his laboratory at the Johnson Foundation, University of Pennsylvania School of Medicine.

Certain preparatory investigations were made in Cold Spring Harbor. From Anderson's previous results, it was clear that T4, the phage used exclusively in experiments described previously in this report, would not be suitable in sonic-vibration experiments, because T4 is more sensitive to destruction by sonics than is the bacterial cell. The smaller phages, T1, T3, and T7, promised to be more suitable, since they are more resistant to sonic vibrations. T3 was chosen for its high resistance and because of the availability of a high-titer anti-T3 serum supplied by Dr. Mark H. Adams. This necessitated preliminary tests with the new phage, using the lysing technique. The latent period of T3 at 37° C. is 13 minutes. To extend this time, the temperature was changed to 30° C., giving a latent period of about 23 minutes. It was found that under these conditions the results of T3 experiments are quite similar to those of T4 experiments. Whether there is any difference between singly infected bac-

teria and those infected by several T₃ particles has not been studied.

One of the most important advantages of the sonic method is that the whole operation of cell disruption can be carried out at low temperatures at which it is impossible to use a procedure involving lysis. It therefore was necessary to know at what temperatures phage growth can be stopped. To determine this, a small aliquot of the source culture (at 30° C.) was placed in a water bath of the temperature to be tested. After 1.5 minutes to allow equilibration, a dilution was made into lysing medium at 30° C. Thereafter, at fixed time intervals, other dilutions into lysing medium were made, and the difference in titer was noted after lysis had taken place. The results are summarized in the following tabulation:

<i>Temperature (° C.)</i>	<i>Time equivalent to 1 min. at 30° C.</i>
3.2 . (no change in 60 min.)	∞
6.3	135
9.6	60
13.3	21
14.9	16

Clearly, little change in phage content of an infected cell takes place at about 3° to 5° C.

The first experiments with sonic vibrations were made to test whether this treatment at 5° C. would liberate T₃ from infected cells during the period when the phage was expected to be present according to the cyanide-lysis experiments. It was found that phage was liberated. Then the optimum length of treatment was determined. It was found that some phage was liberated after 0.5 minutes of treatment, and that approximately the maximum was liberated after 5 minutes of

treatment. If treatment was continued for 60 minutes, the titer of infective centers was reduced to about 50 per cent of the maximum. It was decided to adopt a standard treatment of 5 minutes at 5° C.

The technique used in the lysis experiments was altered by holding dilutions in the lysing medium at 2° C. for 10 minutes in order to give the poison time to penetrate the cell.

Early in the experiments one difficulty arose owing to a phenomenon which, so far as we know, had not been described previously. It was found that the phage-antibody complexes could be broken apart to a detectable extent by sonic vibration. This increased the infective-center count in sonically treated samples where antiserum had been used to inactivate the unadsorbed phage. Therefore it was necessary to remove free phage by differential centrifugation. The technical difficulties in maintaining controlled low temperatures during the centrifugations made accurate prediction of the latent period impossible. The experiments are valid within themselves, however, and the conclusions are based only on intra-experiment comparisons.

As in previously described experiments, a culture (the source culture) containing infected bacteria relatively free of unadsorbed phage particles was used as the source of all subsequent cultures in a given experiment. The titer was about 10⁸ infected cells per milliliter. To make comparisons between material treated with sonic vibrations and material treated with cyanide plus T₆, a dilution of 1:20 was made into GR at 1° to 2° C. The culture was held at this temperature for a short time until the vibrator could be made ready to receive a sample. Then a further dilution of 1:20 was made simultaneously into the vibrator cup and into lysing medium at 2° C. A plating was made

immediately from the vibrator cup before starting the treatment; this constituted the control plating. The standard treatment of 5 minutes' vibration at about 5° C. was given, and followed by plating from the treated material. The sample in the lysing medium was held at 2° C. for 10 minutes and then placed in a water bath at 30° C.

fact that the sonic curve is approximately 1 minute to the right of the CN⁻ + T6 curve. It seems probable that the true intracellular phage picture lies between the two curves. The sonic treatment would be expected to give a lower count than the true count for two reasons: (1) not all the infected bacteria are broken down by

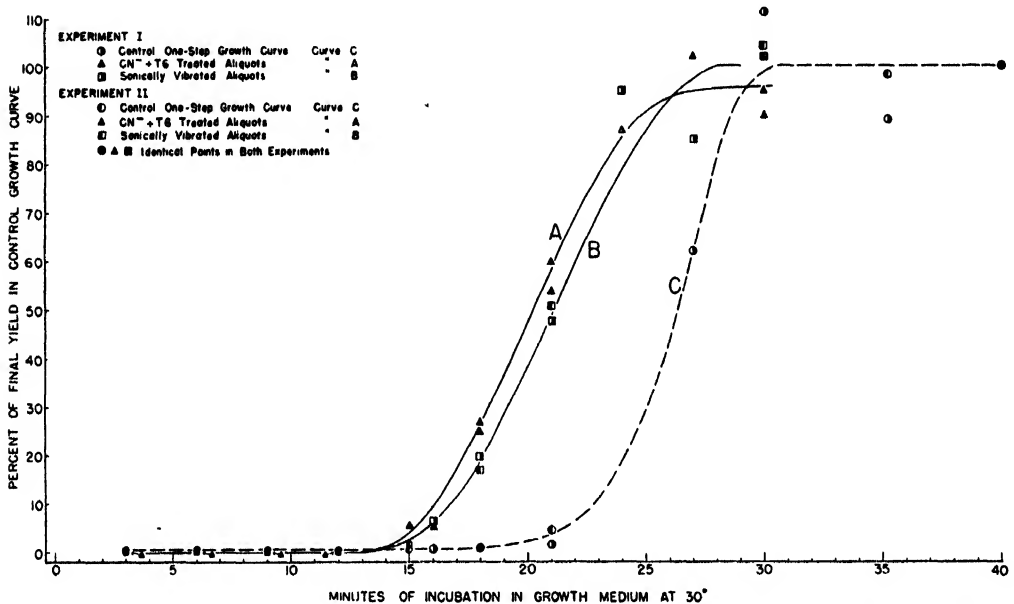


FIG. 4. Comparison of the cyanide-lysis procedure with the sonic-vibration procedure for estimation of intracellular phage. Description of the methods of obtaining these points is given in the text.

After 60 minutes at 30° C., a plating was made from it. From these three sets of platings, comparable determinations could be made for (1) the control growth curve, (2) the sonic estimation of intracellular phage, and (3) the CN⁻ + T6 estimation of intracellular phage. Results from two experiments made in an identical manner are given in figure 4.

The results show fairly close agreement between the two methods of estimating intracellular phage. There does appear to be a consistent difference, shown by the

the treatment, but only about 95 per cent; and (2) a small amount of phage inactivation is to be expected with this treatment. On the other hand, it is not unlikely that the other curve is slightly too high, because the poisoning effect may not be immediate and may not necessarily inhibit the terminal reaction in phage synthesis.

It is hoped that the techniques described can now be utilized to investigate the kinetics of phage growth from both a biochemical and a genetic standpoint.

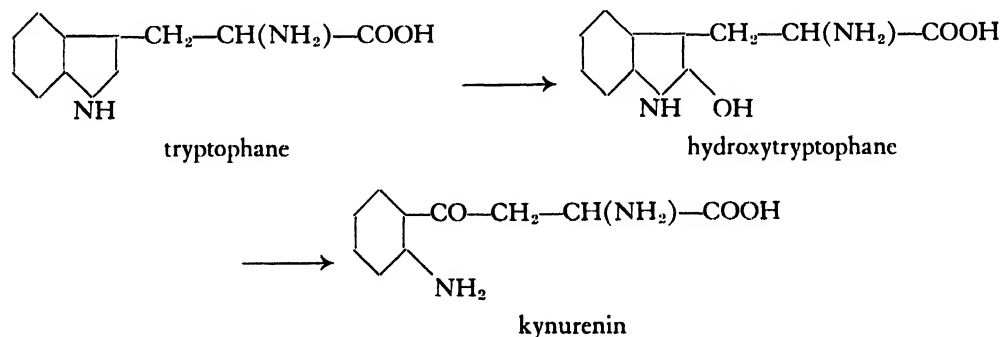
GENIC ACTION

E. CASPARI AND J. RICHARDS

Intensive study has been made in our laboratory of the action of a small number of individual genes. For this purpose, a gene *a* (red eyes) in the meal moth *Ephestia* and a group of pseudoalleles in the mouse, *T*, *Ki*, and *Fu* (short and kinky tail), were selected. In this work we were considerably aided by advice received from Dr. Margaret R. McDonald. During the summer months, we were assisted by Miss Erida Reichert and Mr. Robert H. Tamis; and Dr. and Mrs. Melvin M. Green worked in our laboratory on related problems concerning eye-color genes and pseudoalleles in *Drosophila*.

THE GENE *a* IN EPHESTIA

The gene *a* in *Ephestia kühniella* is a recessive mutant inducing the production of red instead of black eyes and suppressing a number of other pigmentation characters. It has previously been found that the reduction of pigments is due to the inability of *aa* animals to produce kynurenin. In the mammalian organism kynurenin is formed from tryptophane according to the reaction:



converted in this manner is stored in *aa* animals, in part at least in the proteins, so that it must be assumed that qualitatively different proteins are formed in the two strains.

Determination of tryptophane. Part of our work was concerned with a more thorough investigation of the colorimetric methods for the determination of tryptophane. These methods are reliable in purified proteins, but in extracts from whole animals related substances may be expected to interfere. The possible interference of indol, skatol, indolbutyric acid, indolpropionic acid, indolacetic acid, tryptamine, kynurenin, and anthranilic acid with three different colorimetric reactions was investigated. Of these, Eckert's method proved to be least specific, giving the same color reaction with tryptophane, kynurenin, and anthranilic acid. The Shaw-McFarlane method using glyoxylic acid proved to be most specific. Kynurenin and anthranilic acid do not interfere with this reaction, and the other substances, except tryptamine, give color shades different from that produced by tryptophane; their

It was assumed that in *aa* tissues the conversion of tryptophane to kynurenin is blocked. Some of the tryptophane not

presence can therefore be detected by the use of different color filters. On the other hand, substances usually present in un-

purified biological extracts, such as sugars and nucleic acids, produce a brown color with this method, which may obscure the purple color due to tryptophane. For this reason, consistently lower values for tryptophane were found by this method in the same samples of extracts of *Ephestia* than by modifications of the Bates method using *p*-dimethylaminobenzaldehyde. With this latter method, kynurenin and anthranilic acid do not interfere, whereas the other substances mentioned form a color that fades more rapidly than that produced by tryptophane; small amounts of these substances would therefore not interfere with the measurement of tryptophane. All substances mentioned, except kynurenin and tryptophane, can be removed by extraction with ether and toluene. Since kynurenin does not interfere with the Bates method, measurements after extraction with organic solvents are a reliable indication of the amounts of tryptophane present. Dr. Green, with the advice of Dr. A. H. Doermann, worked out a microbiological method for the determination of tryptophane, using a tryptophane-requiring strain of *Escherichia coli*. The microbiological method is about five times more sensitive than the colorimetric ones, measuring amounts as low as 2.5 μ g. of tryptophane. Unlike the Bates and Shaw-McFarlane methods, it is applicable to non-protein tryptophane only and therefore requires hydrolysis of the proteins. Results obtained by this method with extracts of *Drosophila* check well with values obtained for the same samples by the Horn-Jones modification of the Bates method.

Influence of the gene a on protein constitution of Ephestia. The larval proteins of a^+a^+ and aa *Ephestia* were investigated in strains made reasonably isogenic by 7 to 9 generations of outcrossing. The question was investigated whether the increased amount of tryptophane in aa ani-

mals is restricted to one particular protein fraction, or whether it is distributed over a large number of proteins. Larvae of a^+a^+ and aa constitution were ground up in a Waring Blender in 0.7 per cent saline solution. After centrifugation, the dissolved proteins were fractionated by gradual addition of ethyl alcohol. In this way six fractions were obtained, precipitated at concentrations of 16.5, 28.5, 37.5, 44.5, 50, and 75 per cent ethyl alcohol, respectively. These fractions were con-

TABLE 1
PERCENTAGE TRYPTOPHANE IN SEVEN PROTEIN
FRACTIONS OF a^+a^+ AND aa EPHESTIA

Strain	Percentage in fraction						
	1	2	3	4	5	6	7
a^+a^+ ..	1.98	1.86	2.08	2.64	2.80	1.64	2.77
aa	2.70	2.10	2.04	2.71	3.08	1.88	2.77

sistently different in their solubility in 5 per cent NaOH and in their tryptophane content. A seventh fraction was obtained by extracting the residue of the saline extraction with N/20 NaOH at 0° C. and precipitating the proteins with trichloroacetic acid. All precipitates were dissolved in 5 per cent NaOH. In these seven solutions, tryptophane was determined by the Sullivan-Hess modification of the Bates method, and nitrogen by a microkjeldahl procedure. The results of these determinations are shown in table 1. Tryptophane is expressed as percentage of protein, the protein value being obtained by multiplying the N-value by 6.25. The values represent the means of 9 determinations each for a^+a^+ and aa .

In fractions 3, 4, and 7, the two strains contain equal amounts of tryptophane, whereas in fractions 1, 2, 5, and 6, the tryptophane content is higher in aa than

in a^+a^+ material. The difference, however, is not large enough to be statistically significant in any one of the fractions, so that no definite conclusions can be drawn from this experiment.

The behavior of a^+a^+ and aa proteins under the influence of proteolytic enzymes was also investigated. Isogenic a^+a^+ and aa larvae were ground up at 0°C ., in homogenizers as described by Potter and

proteolytic enzymes or in the protein substrate. In either case it would confirm the previous conclusion that there is a difference in the protein make-up of a^+a^+ and aa cells. No difference was found between the two strains for the rate of release of tryptophane in relation to the release of nitrogen.

On the basis of these results, an alternative explanation of the action of the gene a may be suggested. The primary action of the gene a may be to induce the production of qualitatively different proteins. These proteins are richer in tryptophane and their rate of breakdown is slower than is the case in a^+a^+ cells. Therefore, less free tryptophane will be available in aa animals for kynurenin formation and consequently for pigment production.

Tryptophane pyrrolase in Ephestia. Since kynurenin in the mammalian metabolism is formed from tryptophane, experiments have been performed to demonstrate an enzyme catalyzing this reaction in *Ephestia*, and to find differences in the activity of this enzyme between a^+a^+ and aa material. An enzyme of this type is called tryptophane pyrrolase (Kotake *et al.*, *Ztschr. f. physiol. Chem.*, vol. 270, pp. 83-85, 1938) because it causes opening of the pyrrol ring of the tryptophane molecule.

Attempts to demonstrate tryptophane pyrrolase in *Ephestia* material were essentially negative. Homogenates of a^+a^+ and aa *Ephestia* larvae and extracts in Ringer solution and water obtained by centrifugation of homogenate were observed in Warburg manometers with and without addition of $l(-)$ -tryptophane. At the end of the experiment, proteins were removed by precipitation with trichloroacetic acid, and in the filtrates tryptophane was determined by the Sullivan-Hess modification of the Bates method, and kynurenin by the method of Otani and Nishino. In homog-

TABLE 2

NONPROTEIN NITROGEN (NPN) AND NONPROTEIN TRYPTOPHANE (NPT) RELEASED IN AUTOLYSIS AT 31°C . FROM a^+a^+ AND aa HOMOGENATES
(NPN and NPT expressed as per cent of total N and total T)

Time (hrs.)	NPN (per cent)		NPT (per cent)	
	a^+a^+	aa	a^+a^+	aa
0.	10.9	10.2	3.2	6.3
1.	17.7	15.5	7.6	9.1
2.	20.7	17.4	10.0	9.4
4.	26.3	19.7	14.7	12.1
6.	28.7	21.1	16.7	13.0

Elvehjem, in a Ringer solution buffered to pH 6.8. The homogenates were kept in a water bath at 31°C . for 6 hours. From samples taken out at 0, 1, 2, 4, and 6 hours the proteins were removed by precipitation with trichloroacetic acid. The samples were filtered, and nonprotein nitrogen and nonprotein tryptophane were determined in the filtrates. In table 2, the mean values for released nonprotein nitrogen and nonprotein tryptophane from 8 a^+a^+ samples and 7 aa samples are expressed as percentages of the total nitrogen and tryptophane present in the samples.

The results indicate a faster rate of autolysis for a^+a^+ than for aa proteins, as indicated by liberation of both nitrogen and tryptophane. It cannot be decided whether this is due to a difference in the

enates, O_2 consumption frequently was increased after addition of tryptophane to both a^+a^+ and aa material. This increase in O_2 consumption, however, did not occur in all experiments. It appears to be correlated with the formation of a black pigment which is chemically different from the ommochrome pigments derived from kynurenin. In experiments with homogenates, the tryptophane content of the solutions increased, owing to the autolysis described above, whereas no kynurenin could be demonstrated. When extracts from *Ephestia* larvae were used, no increase in O_2 consumption or formation of kynurenin could be observed, whereas the amount of tryptophane was consistently decreased, suggesting the breakdown of tryptophane by a different pathway. Similarly, in experiments carried out with Dr. Green, no kynurenin could be demonstrated after addition of tryptophane to homogenates of wild-type and vermilion *Drosophila*.

Our method for the demonstration of tryptophane pyrrolase has yielded accurate results in experiments with homogenates of mouse liver. In these preparations, O_2 consumption, disappearance of tryptophane, and formation of kynurenin could be quantitatively determined. In three independent experiments it was found that 1.2, 1.3, and 1.7 mole O_2 were used to convert 1 mole tryptophane into kynurenin; the theoretical expectation would be 1.5 mole O_2 per mole tryptophane. The enzyme was also demonstrated in extracts from mouse liver, and in the globulin fraction obtained by fractionated addition of $(NH_4)_2SO_4$.

The negative results of our experiments with *Ephestia* homogenates and extracts may be due to the formation of an inhibitor, or to destruction of the enzyme or of a cofactor in grinding up *Ephestia* material. The presence of an inhibitor for

tryptophane pyrrolase from mouse liver was actually demonstrated in *Ephestia* homogenates. Addition of *Ephestia* material to mouse-liver homogenates inhibited the oxidation of tryptophane to kynurenin. In quantitative experiments it was found that there was no difference in inhibitory activity between a^+a^+ and aa *Ephestia* homogenates, and that the amount of *Ephestia* material necessary to produce observable inhibition was about one-fourth the amount of the mouse material.

Attempts to obtain evidence for the existence of tryptophane pyrrolase in a^+a^+ *Ephestia* material by extraction of the material in H_2O , buffer solutions, and glycerine; by the use of different concentrations of *Ephestia* homogenates, including dilutions of 1:40; by fractionated precipitations of *Ephestia* extracts with $(NH_4)_2SO_4$; or by variations of the temperature and of the pH gave consistently no indication of formation of kynurenin. Therefore attempts were made to demonstrate formation of kynurenin from tryptophane in living *Ephestia* organs rather than in cell-free extracts and homogenates.

It has been shown previously by transplantation experiments that larval testes of a^+a^+ *Ephestia* release relatively large amounts of kynurenin. It can be estimated that one testis releases about 1.5 μg . of kynurenin in 24 hours. Testes were dissected out of full-grown a^+a^+ and aa *Ephestia* larvae and observed in the Warburg in a neutral Ringer solution isotonic for *Ephestia* at 25° C., with and without addition of $l(-)$ -tryptophane. The number of testes and the time of observation were adjusted in such a way that the amount of kynurenin formed should have been easily demonstrated by the Otani-Nishino method. No difference in O_2 uptake between experimentals and controls was found. No kynurenin was

formed, and the amount of tryptophane did not decrease significantly in either strain. Therefore, although it is established that testes form kynurenin *in vivo*, they do not do so *in vitro* upon addition of tryptophane. Though it cannot be excluded that the conditions *in vitro* have inhibited the transformation of tryptophane into kynurenin by *a⁺a⁺* testes, the possibility cannot be neglected that in *Ephestia* kynurenin is formed from a substance other than tryptophane.

Chemical constitution of granules in Ephestia eyes. Another aspect of pigment formation in *Ephestia* eyes has been studied by cytochemical methods. We are indebted to Miss Helen Gay for aid and advice in these experiments. During development, the pigment is deposited on the surface of granules that can be demonstrated with iron hematoxylin and with Giemsa stain. These granules stain pink with pyronin, and disappear after treatment with purified ribonuclease. The conclusion appears justified that these granules contain ribonucleic acid. In a mutant *wa* in *Ephestia*, which has not been available to us, these granules are not formed, and consequently no pigment is deposited in the eye (Hanser, *Ztschr. f. indukt. Abstamm.- u. Vererbungsl.*, vol. 82, pp. 64-97, 1948). It must be concluded that the gene *wa* interferes with pigment formation by inhibiting the formation of a definite type of granules rich in ribonucleic acid.

PHYSIOLOGICAL GENETICS IN A SERIES OF PSEUDOALLELES IN THE MOUSE

In the mouse, three genes with closely similar phenotypic effects were studied, *T*, *Fu*, and *Ki*. *T* and *Ki* are lethal in homozygous condition, but *Fu* is viable. All three affect the shape and length of the tail and have been shown previously

to be closely linked. For our experiments, these mutants had been previously outcrossed for 16 to 21 generations to a closely inbred Bagg albino strain. The three genes may therefore be considered to have been observed in identical genetic backgrounds, except for genes closely linked to them.

Maternal effect of the gene Fu. For the gene *Fu*, Reed (*Genetics*, vol. 22, pp. 1-13, 1937) described a peculiar maternal effect. The number of phenotypically Fused offspring was larger in matings of normal females to Fused males than in the reciprocal cross. Extensive data obtained in our own colony confirm this observation.

This reciprocal difference was interpreted by Reed as being due to differences in penetrance of the gene *Fu* in reciprocal crosses, since he found that some phenotypically normal animals proved in breeding experiments to be genetically *Fu/+*. After the existence of such "normal overlaps" had been confirmed, the question was investigated whether the differences between the reciprocal crosses were due to differential formation of these overlaps. To test this hypothesis, phenotypically normal animals from crosses of Fused females to normal males and normal females to Fused males were bred to Bagg albino animals. Animals that produced at least one Fused offspring were counted as overlaps. Animals that produced at least 15 normal and no Fused offspring were considered genetically *+/+*. From most of the tested animals, however, over 30 offspring were obtained. Some of the test matings were carried out by Miss Jane Trent at Wesleyan University.

From crosses of Fused females by normal males, 48 normal offspring were tested. Of these, 9 (18.8 per cent) proved to be genetically Fused. This value is lower than the value expected from the ratio obtained in the crosses (28 per cent), but not significantly so. From the reciprocal

cross, 19 normal animals were progeny tested, and 4 (21.1 per cent) proved to be genetically Fused. This value is not different from that obtained for progeny of crosses of Fused females by normal males, and appears to be higher than the value expected from the ratio obtained in these crosses (9.1 per cent). Owing to the small number of individuals tested, this difference from expectation is not significant. These results, therefore, give no definite

TABLE 3

PROGENY FROM BAGG ALBINO FEMALES INJECTED
WITH FUSED ORGANS AND THEIR UNTREATED
LITTER MATES, MATED TO *Fu*/+ MALES

Progeny	Fused	Normal	Per cent Fused
Treated.....	18	27	40.0
Control.....	50	59	45.9

ground for assuming that factors other than differential penetrance are responsible for the differences in Fused offspring from reciprocal crosses.

The maternal effect of Fused mothers may be assumed to have arisen through the production in Fused animals of substances inhibiting the expression of the gene *Fu*. In order to test this hypothesis, Bagg albino females were injected with emulsions of liver from Fused animals. They were then bred to *Fu*/+ males. As controls, untreated litter mates of the injected females were bred to the same males. The results are shown in table 3. The differences between treated animals and controls are not significant.

Serological investigations on the gene T in the mouse. Further investigations on the mouse were intended to demonstrate antigenic differences that can be related to one particular gene. For this purpose

a strain containing the dominant gene *T* (Brachyury, short tail) was outcrossed for 16 to 18 generations to a closely inbred Bagg albino strain containing the normal recessive allele of *T*. After this number of outcrosses it may be assumed that Brachyury animals obtained from the outcrosses are reasonably isogenic with the Bagg albino strain, except for the gene *T* and some genes closely linked to it. As antigen, sperm obtained from the epididymides and vasa deferentia of killed males was used. Sperm suspensions in saline containing 10 to 20 million sperm were injected intraperitoneally into a mouse of a different strain, C₃H. After seven injections, the mouse was killed and the serum obtained. It was tested for agglutination of sperm from a *T* male, and showed a titer of 1:200. It also agglutinated sperm from the Bagg albino strain up to a similar titer. The remainder of the serum was absorbed with Bagg albino sperm. The absorbed serum did not react with Bagg albino sperm, but gave a definite although weak agglutination with sperm from a *T* animal. Since sperm from a *T*/+ animal consists of *T* and + sperm in equal proportions, the serum would not be expected to agglutinate more than half of the sperm present; and this would appear as a weak agglutination. In this experiment, an antigen was demonstrated that is associated with the gene *T* or with genes closely linked to it. In a similar experiment involving the dominant gene *Ki*, no agglutination was observed after the serum was absorbed with Bagg albino sperm. More antisera were obtained, but could not be tested.

A mouse disease affecting only males. Our serological experiments were interrupted by a violent disease, which killed off the great majority of the mature males without affecting the females. The females

continued for three weeks to give birth to healthy litters. Young males were not affected, but became sick on reaching maturity.

The disease was certainly not transmitted by mating, since males kept singly for serological purposes also succumbed to it. As an explanation, an infection or some toxic agent must be considered. The latter assumption appears less likely, because the food was identical with that used by MacDowell, in whose mouse colony no disease occurred.

Dissection of animals near death showed inflammation of the epididymides and vasa deferentia. Furthermore, the kidneys were pale. In animals that had been sick for some days without dying, the testes and vasa deferentia were reduced in size, the kidneys were almost white, and the medulla oblongata was softened. Males that survived the sickness were sterile. A pathological investigation of chronically sick animals was carried out through the courtesy of Professor Lymann Duff and Dr. F. Clarke Fraser, of McGill University. They found marked nephrosis of the kid-

ney and some demyelination of the medulla.

From the affected organs, particularly from inflamed epididymides, vasa deferentia, and kidneys, a number of microorganisms were isolated. Dr. Albert Kellner, of the Biological Laboratory, aided us in identifying the organisms. At least three different species of bacteria were found: a slightly hemolytic streptococcus, a staphylococcus, and an apparently undescribed bacterium. This organism is a nonmotile gram-negative rod, which grew at 20° and 37° C., did not ferment any sugars but liquefied gelatin, and formed on agar a nondiffusible yellow pigment. It remains doubtful whether the organisms isolated were causative agents of the disease or secondary invaders.

It was decided to discontinue investigation of the disease, in order to avoid endangering MacDowell's mouse colony. For the remainder of the year, the mouse colony was not in a state permitting continuation of the serological experiments. It was possible, however, to eliminate the disease and start rebuilding the colony.

MOUSE LEUKEMIA

E. C. MACDOWELL, M. J. TAYLOR, AND L. LEWIS

FREING LINE I FROM A CONTAMINATING VIRUS

The isolation of a virus-like pathogenic agent from a transplanted leukemia (line I) was reported a year ago (Year Book No. 46). Filtrates from 18 of the 204 transfers from no. 1491 to no. 1694 of line I have been tested, and in every case the "virus" was obtained. On the other hand, the "virus" has been eliminated from the transplanted leukemia in a branch of this line, Ib, so that it is possible to study the influence of the "virus" upon the behavior of leukemic cells under various conditions

and to answer certain of the questions posed at the end of the previous report.

To confirm the classification of this filterable agent as a virus, a Seitz filtrate of a line-I leukemic spleen (transfer no. 1616) was used to start a series of 12 subpassages. All the 58 mice used as hosts in this new series showed the characteristic sickness, and all but those sacrificed as donors recovered.

Since there was no indication of any loss of potency during these subpassages, it may be concluded that the agent reproduces itself. The twelfth subpassage was made with a Seitz filtrate of minced spleen

suspended in broth; samples of this filtrate were fully tested for bacteria and found negative.

The previously reported findings that the virus regularly induces resistance to itself and that this resistance does not restrain the activity of subsequently inoculated leukemic cells (100 per cent in 45 mice in 15 independent experiments) suggested the method that has been successfully used to free the leukemic cells from the virus. That the virus could be eliminated by passing the leukemic cells through virus-resistant hosts was far from certain, because the virus could be obtained from spleens of mice that had developed resistance to reinfection. This was true for a month after virus infection, and perhaps longer. The actual procedure employed in freeing the leukemic cells from the virus was to pass line-I leukemia (known to be carrying the virus) through four successive hosts that had recovered from virus sickness induced by cell-free filtrates given 55, 117, 128, or 145 days previously. The dose for each of these four transfers and for the following one to a normal host was very dilute (3×10^2 cells). After six more transfers through normal hosts, using larger doses (5×10^6 to 8×10^7 cells), a test for the virus was made by inoculating two mice with a massive dose (1.0 cc.) of a Pyrex-fine filtrate of a spleen suspension. No sickness or weight loss appeared. These two mice were infected with a Seitz filtrate of a dilute (1:3000) spleen suspension from a branch of line-I leukemia that had not been passed through virus-resistant hosts; in the second week the virus sickness and weight loss were fully manifest. Thus a large dose of the filtrate in question induced neither evident virus sickness nor virus resistance that could be detected by a dilute virus-containing filtrate. Similar tests were made after 19 and 35 passages

through normal hosts, with similar evidence of absence of the virus. This virus-free branch is called line Ib.

FOREIGN HOSTS (BALB)

In the foregoing account the mice were all from strain C58, the strain from which line-I leukemia originated in a spontaneous case. For some years inoculation with line I has killed hosts of the Bagg albino strain (Balb); but the terminal condition has not been leukemia, either in gross or in sections. Recently, unfiltered line I (transfers nos. 1686, 1694, and 1700, known still to carry the virus) was given in massive doses to 9 Balb hosts, 8 of which died in about four days, with conspicuous hyperemia of abdominal wall and genitals, blood in intestine, and involuted thymus, but very slight enlargement of lymphoid tissues. Cell-free filtrates from line I induced violent sickness in Balb hosts, but 12 out of 16 recovered.

In contrast with this, the virus-free branch, line Ib, given in unfiltered, massive doses to 18 Balb hosts, neither called forth any gross signs of sickness or of leukemia, nor caused a death. Of these mice, 14 were subsequently inoculated with the virus-carrying line I and showed the clinical signs of virus sickness but did not die; the other 4, given a Seitz filtrate of line I, developed virus sickness, and 1 died. Thus Balb mice are apparently resistant to the leukemic cells alone, and they usually survive infection with the virus alone; but the combination of virus and leukemic cells has killed them regularly. It is evident that inoculating Balb mice with uncontaminated leukemic cells increases their natural resistance to the leukemic cells alone, so that the combination of leukemic cells and virus acts as the virus alone. Balb mice thus become valuable as tests for the presence of a possible

subclinical virus infection in the supposedly virus-free line Ib. These mice respond much more violently than C58 mice to the virus, and their resistance to leukemic cells makes it possible to give much larger and more inclusive test doses than is possible with filtrates. Such tests were made with Balb hosts on the following transfers of line Ib, counting from the first transfer after passage through the virus-immune hosts: 47th, 51st, 55th, 63d, and 70th. In each case Balb hosts were subsequently infected with the virus, either together with line-I cells, or in Seitz filtrates. In every instance, absence of the virus was indicated both by the absence of clinical response to the Ib inoculation and by the absence of any resistance to the virus sickness after virus infection.

INFLUENCE OF THE VIRUS ON THE LEUKEMIC PICTURE

Comparison of the virus-contaminated branch of line I with the virus-free branch, Ib, within the same calendar period shows that the virus influenced the results even though all the animals died from leukemia 2 or 3 days before the virus alone would have manifested any clinical signs. Although virus alone is virtually non-lethal, when combined with line-I leukemic cells it tends to reduce the average interval before death by increasing the proportion of early deaths. The mean interval (dose, 2×10^7 cells) was reduced from 4.729 to 4.350 days ($\text{Diff.} = 0.379 \pm 0.062 = 6 \times \text{S.E.}$). The terminal spleen length was also reduced; in 167 mice dying from leukemia plus virus, the mean spleen length was 2.656 cm., which was smaller by 0.382 ± 0.018 cm. ($= 21 \times \text{S.E.}$) than the mean for 203 leukemic spleens without virus. The proportion of spleens measuring 3 cm. or more was 70 per cent for line Ib (virus-less) and 2 per cent for line I (with virus). This difference in spleen length was inde-

pendent of the time of death. Although the size of leukemic spleens is less when the virus is present, the virus alone increases the size of normal spleens. This appeared in a series of 6 C58 mice infected with a Seitz filtrate containing the virus, which were killed in the second half of the fifth day, when leukemics would be dying, and were compared with 6 litter mates of the same sex and weight. Lengths of normal spleens ranged from 1.60 to 1.70 cm.; virus spleens ranged from 1.90 to 2.30 cm.; virus spleens averaged 1.26 times the average of the normals. By weight, the normal spleens averaged 100.1 mg.; the virus spleens averaged 244.1 mg., or more than twice as heavy. Weights of the four axillary nodes (weighed together) from the virus mice averaged 1.9 times those from the controls. Thymus weights showed a tendency toward reduction with the virus, although the ranges overlapped and the average with virus was 0.8 times normal. Similar comparisons between groups of 4 virus-infected and 4 normal animals killed after 10 days gave average weights for the virus spleen of 2.5 times normal, for the virus axillary nodes of 3.7 times normal, and for the virus thymus of 0.37 times normal. The observations on the fifth day make it clear that the mouse responds to the presence of the virus before this becomes evident from clinical signs.

Other features of the line-I autopsies that have been either reduced or eliminated by the removal of the virus contamination include: peritoneal ascites, hyperemia of leukemic infiltrations of abdominal muscles and genital organs, and small lung hemorrhages.

VIRUS AND INDUCED RESISTANCE TO LEUKEMIA

The observation that resistance to the virus does not resist leukemic cells made

it clear that the induced resistance to leukemia we have long held in question is either specifically different or dependent upon a different kind of mechanism. But even so, it is possible that the presence of the virus in many of the experiments we have carried out may have considerably altered the results and confused the interpretations. To validate this work, the repetition of certain of these experiments without virus contamination has become a critically important undertaking. The method most extensively employed in this laboratory for inducing resistance to line-I leukemia in C58 hosts is treatment with tissue from strain StoLi 3 days before a dilute, but still 100 per cent lethal, dose of the leukemic cells. This may be expected to give resistance in all cases and enable the mouse consistently to withstand subsequent doses of increasing magnitude. Such results depend upon appropriate test dosage, scaled to the current degree of virulence of the leukemic cells. Three lethal dilutions of the virus-free line Ib were used in testing for resistance 3 days after treatment with minced normal liver from strain StoLi. With a test dose of 125×10^4 cells, 4/9 survived; with a test dose of 78×10^8 cells, 9/10 survived; with a test dose of 12×10^2 cells, 10/10 survived. Of 17 survivors reinoculated with doses of 2×10^7 and 5×10^6 cells, all survived. These results confirm the previous conclusions. In one respect these resisting mice differed from those in the earlier experiment, in that none showed the signs of sickness at the end of a week. The virus may be held responsible for the sickness that previously developed in such resisting mice, but it was not responsible for their survival.

A second method of inducing resistance, fully confirmed by old experiments, is treatment with liver of a mouse of the same strain (C58) after it has been firmly

immunized by a series of massive doses of leukemic cells. This transfer of resistance by immunized tissue has been repeated with the virus-free line Ib. Immune liver from a mouse that had been treated with normal StoLi liver, then inoculated after 3 days with a dilute dose of line Ib, followed at intervals by four massive doses, was given intraperitoneally to 14 mice, which were immediately inoculated with line Ib (doses 125×10^4 or 78×10^8 cells). All the 14 controls died, and all but 1 of the test animals survived. Of these 13 survivors, 8 developed enlarged spleens, which approached leukemic proportions and then returned to normal within a week, but at no time did these mice show signs of virus sickness, in their appearance or behavior.

The sickness formerly noted in mice that were resisting inoculated leukemic cells was long supposed to be connected with the process of destroying the leukemic cells, for it had been learned that the process of resistance destroyed leukemic cells after they had multiplied and extensively invaded the mouse, as indicated by the temporary enlargement of spleens. In the present case, the breakdown of very obvious leukemic enlargements, without any signs of sickness, effectively eliminates the possibility that the former sickness was a reaction to toxic products of leukemic cell destruction.

VIRUS IN OTHER LINES

Seitz filtrates of other lines of transplanted leukemia have been found to transmit pathogenic agents that produce conditions similar to but not identical with the virus sickness of line I. Cross-resistance with the line-I virus indicates basic relationships between these agents, but there are characteristic differences that raise the question of parallel origins or a com-

mon origin with deviations. These virus-like agents have been eliminated from lines L, U, and V by the use of immunized hosts, as in the case of line I, and with similar tests for the success of their elimination. Line L presented a special case, because the ability of this line to induce resistance to itself is so strong that it cannot be transmitted by highly dilute doses, and therefore the presence of a virus cannot be observed clinically by delaying the leukemic death, nor, in freeing the leukemic cells from virus, can advantage be taken of very dilute doses for the immune hosts.

Three cases of spontaneous leukemia have been tested for the virus and found entirely free. The initial susceptibility of every one of the large numbers of mice that have been treated with the virus, and the continued absence of the virus in the four decontaminated lines of transplanted leukemia, constitute excellent evidence that the mice in the breeding colony are virus-free.

A NEW COAT COLOR, LIGHT

A mutation ($B'B'$) from the black hair color of the leukemic strain C58 has produced a strikingly beautiful new coat color, in which the hair tips are "hair brown" (Ridgway) and the rest almost white; eyes are black. Crossed with black, all the first generation ($B'B$) is "chaetura drab" (Ridgway), with the lower quarter of the hairs light gray ("dark"); later generations (F_2 and backcrosses) segregate for black (BB), "dark" ($B'B$), and "light" ($B'B'$), in close agreement with monohybrid ratios (208 mice). "Light" crossed with brown (bb) gives all "dark" ($B'b$) in the first generation, and the following generations segregate for brown (bb), "dark" ($B'b$), and "light" ($B'B'$) in the expected ratios, but no blacks have appeared (474 mice). Thus, this mutation is dominant to black and is in the same chromosome—either in the same locus (allelomorphic), as indicated by the symbols, or closely linked.

GENETIC STRUCTURE OF NATURAL POPULATIONS

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The program of studies on the genetic structure of natural populations that has been carried on for the past ten years (see Year Books Nos. 37-41, 43-46) has used chiefly the small fly species *Drosophila pseudoobscura* and *D. persimilis* as materials for investigation. These are temperate-zone species, native in the western United States, western Canada, and the highlands of Mexico and Guatemala. This work is now to be interrupted for at least one or two years, in order that a study may be undertaken of the population of some species of *Drosophila* in the tropical parts of South America, mainly in Brazil. The justification of this course is that, as will be explained in more detail below, the

genetic population structure in tropical species is likely to be different from that in the inhabitants of temperate climates. The work planned for the coming year is designed to test this hypothesis. Now is an opportune time, therefore, to review the path traversed, and to establish some guideposts for future work.

ADAPTIVE POLYMORPHISM IN POPULATIONS OF *DROSOPHILA*

Populations of some species of *Drosophila* exhibit an interesting variation in the structure of their chromosomes. Owing to the occurrence of inversions of blocks of genes in the chromosomes, several chromosomal types may be present in

a species. Thus, *D. pseudoobscura* has twelve types of third chromosomes, and *D. persimilis* has seven (not counting very rare and strictly endemic types, which may conceivably have originated by recent mutation). Different chromosomal types very frequently occur together in populations of the same locality. Flies carrying chromosomes of different types interbreed at random, so that individuals in which the two chromosomes of a pair are similar in structure (structural homozygotes) and individuals in which the chromosomes of

account for if carriers of certain chromosomal types are better adapted to the environment prevailing at some seasons, and carriers of other types are superior at other seasons. Natural selection would, in this case, augment the frequencies of the chromosome types that have the highest fitness, and depress the frequencies of the types that are relatively less fit, at any given season.

Data on seasonal changes are available for populations of Mount San Jacinto in southern California and for some locali-

TABLE 1

CHANGES OCCURRING IN THE FREQUENCIES OF CHROMOSOME TYPES AT PIÑON FLATS DURING THE SPRING MONTHS OF 1948

Date of collection	ST	AR	CH	TL	SC	Chromosomes examined
April 14, 1948...	62.8	16.8	14.8	5.4	0.2	500
May 23, 1948...	46.0	13.1	28.9	11.8	76
June 3, 1948...	35.6	21.8	35.6	6.9	174

a pair belong to different types (structural or inversion heterozygotes) are encountered.

But flies with all types of chromosomes are externally alike, so that they can be distinguished only by examination of their chromosomes in the cells of the larval salivary glands. This apparent similarity at first led to the supposition that possession of this or that chromosomal type produces no variations in the structure or physiology of the fly's body—that, in other words, the chromosomal types are "neutral characters" as far as natural selection is concerned. This supposition, however, proved to be incorrect (see Year Book No. 40 [1940-1941]). In 1939 and 1940 it was first discovered that populations of *D. pseudoobscura* undergo, in at least some localities, cyclic seasonal changes in the relative frequencies of chromosomal types. Now, such cyclic changes are easiest to

ties in the Sierra Nevada in central California. Less extensive data suggest that such changes occur also in certain localities in Texas. The population of the Piñon Flats locality on Mount San Jacinto has been followed, with some interruptions, since 1939. Standard (ST), Arrowhead (AR), Chiricahua (CH), Tree Line (TL), and Santa Cruz (SC) chromosome types occur in this population. Every spring, the frequency of ST drops from March to June, while the frequency of CH increases. In summer, the reverse change takes place. AR, TL, and SC chromosomes change relatively little. Owing to the kindness of Professor Carl Epling and his students P. Levene and D. Mitchell, samples of *D. pseudoobscura* were collected and sent to this writer in 1948. Examination of these samples disclosed the frequencies of chromosome types shown in table 1.

Despite the high sampling errors, it is clear that ST chromosomes sharply decreased in frequency, and that CH chromosomes increased, within a period of less than two months. In fact, the frequency of ST chromosomes found in the April sample is the highest ever recorded for the Piñon Flats locality, and the frequency of CH the lowest. This exceptional composition of the 1948 samples may conceivably be due to the fact that the winter of 1947-1948 was the driest during the period of our observations. It should be noted that the Piñon Flats locality lies at an elevation of about 4000 feet; some 3200 feet below it, at about 800 feet, lies Andreas Canyon, the population of which normally has a greater frequency of ST and a lower one of CH chromosomes than is present at any given season at Piñon Flats. It may be, then, that the dry and warm winter of 1947-1948 caused a shift of the Piñon Flats population in the direction of the Andreas Canyon population.

The seasonal changes in the Sierra Nevada populations have a character rather sharply different from those on Mount San Jacinto (see Year Book No. 46 [1946-1947]). In the Sierra Nevada, the frequency of ST chromosomes, in midsummer, is greater at lower and less at higher elevations; AR chromosomes show the opposite regularity; and CH, TL, and SC chromosomes change relatively little. Now, at the Aspen Valley (elevation 6200 feet), Mather (4600), and Lost Claim (3000) localities, the frequencies of ST chromosomes increase, and those of AR chromosomes decrease, during the summer; the reverse changes that close the cycle take place, presumably, during hibernation. But the observations made during the summers of 1946 and 1947 failed to disclose significant changes at the neighboring locality of Jacksonville, at an elevation of about 850 feet. Through the

courtesy of Professor C. Epling and his colleagues, the writer received samples collected at Jacksonville in February, March, April, and May 1948. A study of these samples disclosed that during the winter and spring ST chromosomes are less frequent, and AR chromosomes more frequent, than they are in summer (June to September). Thus the population of the Jacksonville locality undergoes seasonal changes similar in kind to those observed at Lost Claim, Mather, and Aspen Valley. These changes are completed at Jacksonville, however, before the advent of the hot summer weather, when they are taking place in the higher localities.

EXPERIMENTAL CONFIRMATION OF THE NATURAL-SELECTION HYPOTHESIS AS APPLIED TO ADAPTIVE POLYMOR- PHISM IN *DROSOPHILA*

The discovery of seasonal changes in the genetic composition of populations of *D. pseudoobscura* has opened up possibilities for experimental studies on the mechanisms of natural selection. Indeed, the changes observed in natural populations are so rapid and so striking that the selective forces involved must be very great. This amounts to saying that the flies that are carriers of different chromosome types must show large differences in fitness. Such large differences must be experimentally detectable.

For this purpose artificial populations were set up in "population cages" in the laboratory, and the changes that took place in the course of time in such populations were recorded (see Year Books Nos. 43-46). It became clear at once that the carriers of different chromosome types in fact possess very different degrees of fitness in populations kept at temperatures around 25° C. Curiously enough, populations kept at 15-16° C. show no changes with time in the proportions of the chromo-

somal types, thus proving that the adaptive values of these types are about alike at that temperature. The relative fitness of the chromosome types is, accordingly, very sensitive to environmental variations.

The population-cage experiments have disclosed another very significant relationship. If the chromosome types introduced into a cage are derived from wild ancestors collected in the same locality in

less frequent among the adult flies than is indicated by the above formulas. (The carriers of the chromosome types are known to interbreed at random, and the differences in fitness are assumed to lead to differential mortality between the egg and the adult stages.) Taking the fitness of the ST/CH heterozygotes to be unity, the best estimates for the homozygotes of Piñon Flats origin are as follows:

$$\text{ST/CH} = 1.00 \quad \text{ST/ST} = 0.81 \quad \text{CH/CH} = 0.47$$

nature, the fitness of structural heterozygotes is higher than that of the corresponding homozygotes. Consider, for example, populations that are mixtures of ST and CH chromosomes derived from flies col-

Similar estimates of the fitness of the different types in population cages containing ST and AR or AR and CH chromosomes of Piñon Flats origin are as follows:

$$\begin{array}{lll} \text{ST/AR} = 1.00 & \text{ST/ST} = 0.81 & \text{AR/AR} = 0.50 \\ \text{AR/CH} = 1.00 & \text{AR/AR} = 0.86 & \text{CH/CH} = 0.48 \end{array}$$

lected at Piñon Flats, in California. Since the flies in such populations interbreed at random, heterozygotes ST/CH and homozygotes ST/ST and CH/CH are produced. Their fitness can be estimated by two methods. First, one observes the rate of change in the frequencies of ST and

The experiments with population cages containing chromosomes derived from the population of Mather, in the Sierra Nevada of California, gave a somewhat different set of estimates. Taking the fitness of the heterozygotes always to be unity, we have the following relationships:

$$\begin{array}{lll} \text{ST/CH} = 1.00 & \text{ST/ST} = 0.78 & \text{CH/CH} = 0.28 \\ \text{ST/AR} = 1.00 & \text{ST/ST} = 0.64 & \text{AR/AR} = 0.58 \\ \text{AR/CH} = 1.00 & \text{AR/AR} = 0.81 & \text{CH/CH} = 0.60 \\ \text{ST/TL} = 1.00 & \text{ST/ST} = 1.12 & \text{TL/TL} = 0.33 \\ \text{AR/TL} = 1.00 & \text{AR/AR} = 0.69 & \text{TL/TL} = 0.12 \end{array}$$

CH chromosomes with time. The degree of fitness is deduced from the regression of the amount of change per generation of the frequency of a given chromosome type at different times. Second, if the frequencies of two chromosome types in a population are q and $(1 - q)$, the two homozygotes must be formed in proportions q^2 and $(1 - q)^2$, and the heterozygotes $2q(1 - q)$. If, however, the fitness of the heterozygotes is higher than that of the homozygotes, the former will be relatively more and the latter relatively

Two facts should be noted particularly about these estimates. First, the differences in the fitness of the different genotypes are often very large. Thus, the CH/CH homozygotes from Piñon Flats and the CH/CH and TL/TL homozygotes from Mather have a fitness less than half that of the ST/CH, AR/CH, ST/TL, and AR/TL heterozygotes. If the fitness of the heterozygotes is regarded as "normal," the homozygotes are to be classed as semi-lethal. Yet such homozygotes occur in natural populations. Second, only one

heterozygote—namely, ST/TL from Mather—has a fitness lower than one of the corresponding homozygotes, ST/ST (1.00 and 1.12, respectively).

EQUILIBRIUM IN POPULATIONS

If heterozygotes for two allelic genes or two chromosome types have a higher fitness than the corresponding homozygotes, natural selection does not lead to elimination or establishment of either of the competing genetic variants. Instead, an equilibrium is reached, at which the competing variants—in our case the different chromosome types—are preserved indefinitely with certain predictable frequencies. The adaptive polymorphism becomes a permanent condition in the population. The biological meaning of such a situation is easy to understand. Adaptive polymorphism permits a species to inhabit diverse environments and to exploit diverse ecological niches. Other things being equal, a polymorphic species has greater adaptive resources than a genetically uniform one. Now, if one genetic variant is superior to another and the hybrid (heterozygote) is intermediate in fitness, the polymorphism is unstable. Suppose, for example, that ST/ST flies were superior to CH/CH in summer but inferior in spring; assume, however, that the heterozygotes ST/CH were intermediate in fitness between the homozygotes, instead of being superior to both as is actually the case. With heterozygotes intermediate, a particularly warm or prolonged summer might lead to a complete elimination of CH chromosomes and the establishment of ST to the exclusion of other chromosome types. A population that had lost CH chromosomes would find itself at a disadvantage the next spring, since in spring CH would be superior to ST. Such a “selective accident” cannot happen if ST/CH heterozygotes

are superior to ST/ST and CH/CH homozygotes, because natural selection will always establish an equilibrium between ST and CH types, and will not eliminate either of them. Superiority of the heterozygotes buffers populations against environmental change.

These theoretical considerations have been confirmed in several ways. With a single exception, in no population cage has any one of the competing chromosome types derived from the same natural population been eliminated; instead, equilibrium has been established and the artificial populations in the cages have remained static thereafter. The exception was found when TL chromosomes derived from the Mather populations competed with ST chromosomes. As shown above, ST/ST homozygotes are superior to ST/TL heterozygotes as well as to TL/TL homozygotes. Accordingly, the process of selection leads to elimination of TL and complete victory of ST chromosomes (see Year Book No. 46). The reason TL chromosomes are retained in the Mather population at all is that they form highly fit heterozygotes with another chromosome type, AR, found in the same population. As shown above, TL/AR heterozygotes are superior to AR/AR and to TL/TL homozygotes.

Conclusive evidence of the adaptive superiority of the chromosomal heterozygotes in nature has been obtained in the following way (in co-operation with Dr. Howard Levene). According to the binomial-square (Hardy-Weinberg) rule, if chromosome types in a panmictic population have frequencies p , q , r , etc., the homozygotes and heterozygotes are formed in proportions $p^2 + q^2 + r^2 + \dots + 2pq + 2pr + 2qr + \dots = 1$. It has been shown that this rule is in fact applicable to natural populations of *D. pseudoobscura*. If wild females inseminated in nature are allowed

to produce offspring in captivity, and if the offspring is grown at optimal conditions, the proportions of chromosomal heterozygotes and homozygotes among the larvae obtained fit the binomial-square rule. The fitness of the homozygotes, however, is supposed to be lower than that of the heterozygotes. Provided that the differences in fitness result in a differential mortality between the egg and the adult stages, heterozygotes should be more, and homozygotes less, frequent among the adult flies found in nature than the binomial-square rule would demand. This hypothesis proved to be difficult to test, because with several chromosome types present in most natural populations, a prohibitive number of flies would have to be examined for chromosome constitution from a single sample of a natural population. In the course of the work, however, many small samples were accumulated, none of which taken singly had much statistical significance. Dr. Levene evolved a statistical technique whereby an aggregate of such individually insignificant samples could be evaluated. With the aid of this technique, it has been shown that chromosome heterozygotes are indeed more frequent, and homozygotes less frequent, in natural populations than they should be according to the binomial-square rule (the probability that the observed deviation may be due to chance is less than 10^{-6}). It can further be estimated that the observed deficiency of chromosome homozygotes is about 15 per cent, which indicates a considerable differential mortality of the homozygotes between the egg and the adult stages.

EXPERIMENTAL MODIFICATION OF THE EQUILIBRIUM VALUES

In a population in which two or more chromosome types are present, natural selection leads toward establishment of an equilibrium. After an equilibrium is

established, no further changes in the frequencies of the chromosome types should occur. Yet we know that seasonal changes do occur in some natural populations. This means that the equilibrium values at which the populations are stabilized can be modified by environmental influences. As indicated above, experiments with population cages have shown that the relative fitness of the chromosome types is indeed extremely sensitive to temperature; whereas at 25° the fitnesses of the heterozygotes and homozygotes are sharply different, they are nearly or completely alike at 15° . The situation at temperatures intermediate between 15° and 25° has not been studied sufficiently, but the data as they stand indicate that only the speed of the selection process, not the equilibrium values, is changed by temperature variations. It was decided to test whether nutritional agents might be more effective in this respect.

Thanks to the courtesy of Professor R. P. Wagner, of the University of Texas, the writer received a collection of five strains of yeast and four strains of bacteria isolated by Dr. Wagner from the crops of wild *D. pseudoobscura* flies caught at Piñon Flats, California, in April 1946 and April 1947. In co-operation with Mr. Stanley Zimmering, experiments were undertaken in which population cages were given culture medium infected with one or another of the yeasts of Dr. Wagner's collection. All other experiments used commercial Fleischmann's yeast for food. Thus far only three experiments have been made with Dr. Wagner's yeasts, and even these had to be discontinued earlier than would have been desirable. Nevertheless, some clear-cut results have been obtained, as shown in table 2.

The initial populations in all cases had about 48 per cent ST and 52 per cent CH chromosomes of Piñon Flats, California, origin. In cage no. 53, provided with yeast

no. IV, the proportions of ST chromosomes rose rather slowly, attaining about 70 per cent in about 7 months from the start of the experiment. This is close to the equilibrium value obtained with Fleischmann's yeast under similar experimental conditions. In cage no. 54 the frequencies of ST rose with significantly greater rapidity, attaining almost 90 per

46) in which chromosomes from one locality (for example, ST from Mather) competed with chromosomes of a different type from a different locality (CH from Piñon Flats) gave significantly different results. Among the adult flies that hatched in such cages, characteristic deviations from the proportions of heterozygotes and homozygotes demanded by the binomial-

TABLE 2

CHANGES IN FREQUENCIES OF CHROMOSOME TYPES OCCURRING WITHIN POPULATIONS FED WITH DIFFERENT STRAINS OF YEAST

TIME	CAGE NO. 52: YEAST I		CAGE NO. 53: YEAST IV		CAGE NO. 54: YEAST V	
	ST	CH	ST	CH	ST	CH
Started November 12, 1947	47 9	52 1	47 9	52 1	.	.
Started December 12, 1947.....	48 0	52 0
Mid-December 1947.....	54 7	45 3	46 3	53 7	.	.
Mid-January 1948	55.3	44 7
Mid-February 1948	72 7	27 3	59 0	41 0	75 0	25 0
Early April 1948.....	62.7	37 3	82 3	17 7
Early June 1948.....	70 3	29 7	89 7	10.3

cent in 6 months. This is considerably above the equilibrium point obtained with Fleischmann's yeast. In fact, it is not certain whether the ST/CH heterozygotes in cage no. 54 had a fitness greater than ST/ST homozygotes, or whether ST chromosomes would eventually have displaced CH entirely if the experiment had been continued longer. Experiments on the influence of nutritional agents on the fitness of the chromosomal types should be resumed at the earliest opportunity.

COMPETITION OF CHROMOSOMES OF DIFFERENT GEOGRAPHIC ORIGIN

In the experiments mentioned up to this point, the competing chromosome types were always derived from the population of a single locality—either Piñon Flats on Mount San Jacinto, or Mather in the Sierra Nevada of California. The first experiments (mentioned in Year Book No.

square rule were observed. These deviations were of a kind which indicated that the heterozygotes carrying two chromosomes of different types from different localities are not superior, but rather intermediate in fitness between the corresponding homozygotes, which in these experiments carried two chromosomes of the same type and of the same geographic origin.

Now, if the fitness of a heterozygote is less than that of one of the homozygotes, natural selection will lead not to any equilibrium, but to elimination of the less fit chromosome type. To test this hypothesis, the five population cages that contained chromosomes derived partly from Piñon Flats and partly from Mather localities were kept for about a year, and the frequencies of the competing types of chromosomes were determined in samples taken from time to time. The progress of

selection in a cage (no. 45) containing AR chromosomes of Mather and CH of Piñon Flats origin is shown in table 3.

The frequency of AR continued to increase, and that of CH to drop, until the end of the experiment, although the rate of change progressively diminished. The diminution is expected to occur when the least fit type in the population—homo-

TABLE 3

CHANGES IN FREQUENCIES OF CHROMOSOME TYPES OCCURRING IN A POPULATION CONTAINING CHROMOSOMES OF DIFFERENT GEOGRAPHICAL ORIGIN (SEE TEXT)

Time	AR	CH
February 3, 1947.....	50.0	50.0
Early March.....	60.7	39.3
Early April.....	61.3	38.7
Early June.....	73.3	26.7
Early July.....	76.3	23.7
Early August.....	82.3	17.7
Early September.....	83.7	16.3
Mid-November.....	88.7	11.3
Late January 1948.....	89.7	10.3
Late March 1948.....	91.0	9.0

zygous CH/CH—becomes rare; the data are consistent with the supposition that the CH chromosomes would eventually be supplanted by AR.

Similar elimination of the weaker competitor tended to occur in the cage (no. 48) containing ST chromosomes of Mather and CH of Piñon Flats origin, and in no. 49, where ST chromosomes of Piñon Flats competed with CH chromosomes of Mather origin. But in cages nos. 46 and 47, in which ST chromosomes from Piñon Flats competed with AR from Mather and vice versa, no chromosome was victorious and equilibria became established. This means that the fitness of the ST/AR heterozygotes was on the whole greater than that of ST/ST and of AR/AR homozygotes, although the viability of ST/AR between the egg and the adult stages was

below that of the ST/ST class. In other words, the handicap that ST/AR heterozygotes have in this case at stages below the adult is overbalanced by an advantage in the adult stage. This shows how complex are the phenomena of natural selection when chromosomes of different geographic origin are brought together in competition.

The high fitness of heterozygotes carrying chromosomes of different types from the same locality may be described as heterosis (hybrid vigor). One may say, then, that hybridization of strains of different geographic origin sometimes leads to loss of heterosis. In view of the high theoretical interest of this phenomenon, some experiments were arranged to study it further. Following a suggestion of Professor H. J. Muller, a procedure was devised that produced, in a population cage, a population consisting at fertilization of 25 per cent ST/ST homozygotes, which carried one ST chromosome of Mather and the other of Piñon Flats origin; 25 per cent AR/AR homozygotes, with one chromosome of Mather and the other of Piñon Flats origin; and 50 per cent ST/AR heterozygotes, which carried the two chromosomes of different types but coming from the same geographic locality (either from Mather or from Piñon Flats). In other words, in this experiment structural homozygotes that were interlocality hybrids competed with structural heterozygotes that had both chromosomes from a single locality. Two population cages of this kind (nos. 50 and 51) were raised with the assistance of Mr. M. Foster. Adult flies developing in these population cages were tested for chromosomal constitution. The combined results obtained are as follows:

ST/AR heterozyg.	ST/ST homozyg.	AR/AR homozyg.	Total tested
246	114	63	423

It can be seen that ST/AR heterozygotes were slightly more than twice as numerous as ST/ST homozygotes, and almost four times as numerous as AR/AR homozygotes. (The "raw" counts shown here must be corrected for undetected heterozygotes, which should constitute $1/31$ part of the detected ones; this technical detail will be discussed in another publication.) The conclusion is that structural heterozygotes having the two chromosomes of a pair derived from the same geographic locality are superior to structural homozygotes that carry the two chromosomes from different localities. Heterosis is caused by heterozygosis for chromosomes of different types but of similar geographic origin. Hence, no heterosis results from heterozygosis for two chromosomes of the same structural type but of different geographic origin.

In the experiments thus far observed, the chromosomes have been derived from geographic localities only some 300 miles apart, and furthermore belonging to the same climatic zone of California montane forests. In two other experiments (cages nos. 55 and 56), the competing chromosomes originated in populations very remote from each other. ST or AR chromosomes from California (Piñon Flats) competed with CH chromosomes from Santa Barbara, Chihuahua, Mexico (the latter coming from a sample of flies collected by Professor H. Spieth). F_1 generation hybrids of ST flies of California origin and CH flies of Mexican origin were introduced in cage no. 55. Among the F_2 generation flies developed under the stringent competitive conditions prevailing in population cages, 260 individuals were taken and their chromosomal constitution determined. The following distribution of heterozygotes and homozygotes was found:

Heterozygotes	Homozygotes	Homozygotes
ST/CH	ST/ST	CH/CH
122	81	57

If the heterozygotes and homozygotes are equally viable, the heterozygous class should be twice as numerous as each homozygous class. Heterosis would make heterozygotes more than twice as numerous. In reality, ST/CH heterozygotes are distinctly less than twice as frequent as ST/ST homozygotes, and about twice as common as CH/CH homozygotes. There is, accordingly, no heterosis; the viability of the heterozygotes is about equal to that of the less viable homozygous class.

In population cage no. 56, the initial population consisted of F_1 flies from the cross AR from California to CH from Mexico. Among the F_2 flies that developed in the cage, 252 were analyzed for chromosomal constitution, with the following results:

Heterozygotes	Homozygotes	Homozygotes
AR/CH	AR/AR	CH/CH
104	84	64

Here the heterozygotes seem to be less than twice as frequent as *either* homozygous class. Among the experiments thus far performed, this is the first in which the chromosome heterozygotes are not superior to at least one of the homozygotes. It follows that heterozygosis for chromosomes of remote geographic origin not only leads to no hybrid vigor, but may lead to inferiority of the heterozygotes.

HETEROSIS AND COADAPTATION

It is clear that heterosis is not a necessary consequence of heterozygosis for two different chromosome types. Such heterozygosis produces hybrid vigor when the two chromosomes are derived from the same population (with the exception of the ST-TL case discussed above). When the chromosomes come from different but geographically not very remote populations, the results are irregular; the rule appears to be that the heterozygote is intermediate in fitness between the homo-

zygotes, but some slight heterosis is occasionally produced. Finally, if the chromosomes are derived from very distant populations, there is no hybrid vigor and there may be some deterioration.

Two conclusions follow from these experiments. First, chromosomes of the same type may carry different gene complexes in geographically different populations. (This conclusion is arrived at on the basis of independent evidence if we compare the changes taking place in population cages that contain the same competing chromosome types from one locality, such as Piñon Flats, with those in cages with chromosomes from another locality, such as Mather; see above.) Secondly, the gene contents of the chromosomes of different types in any one geographic locality are mutually adjusted to each other in such a way that a high fitness is produced in the heterozygotes. This mutual adjustment, or coadaptation, of the gene contents of the chromosomes of different types is evidently the result of a process of natural selection. Since in many natural populations of *D. pseudoobscura* chromosomal heterozygotes are more frequent than homozygotes, the process of coadaptation results in an increase in the level of fitness of the population as a whole. But the process of coadaptation has taken place independently in geographically remote populations. As a consequence, no increase, or even a decrease, in fitness results from hybridization and heterozygosis for chromosomes derived from different populations.

Several problems of considerable interest, which must be studied as soon as possible, arise in connection with the above facts. First of all, geographic regions exist (parts of Arizona, New Mexico, Utah, and Colorado) in which populations of *D. pseudoobscura* are predominantly or en-

tirely homozygous for a single chromosome type, namely, AR (Arrowhead). Homozygotes for AR chromosomes of California origin have so low a fitness that it seems unlikely that they would constitute a natural population of so extensive a territory. It is more likely that AR chromosomes of the nonpolymorphic territory differ radically in their properties from those found in California. Possibly, a highly fit homozygous type has evolved.

As pointed out in Year Book No. 46 (p. 163), the divergence of the gene contents of the chromosomes in different populations may lead to splitting up of an originally single species into two or more derived ones. Suppose that a territory inhabited by populations with a certain set of coadapted gene complexes abuts on a territory with a different set of coadapted complexes, but that no intermediate gene complexes exist. If the gene complexes of the two territories give a low fitness in the heterozygotes (race hybrids), a genetic rift has occurred in the species along the geographic boundary between the two territories. But no such rift will be present if chains of intermediate gene complexes, each tolerably coadapted with respect to the complexes in the neighboring populations, are formed. Populations of *D. pseudoobscura* in California and neighboring states seem admirably fitted for investigation of this type of problem.

ADAPTIVE EVOLUTION IN DIFFERENT ENVIRONMENTS

The biological significance of adaptive polymorphism is that, as pointed out above, it permits a species to exploit the accessible environments more fully than a monomorphic species could do. Now, the range of environments with which an organism comes in contact differs not only

for different species but also for populations of the same species. Other things being equal, species that inhabit temperate and frigid zones, or tropical climates with great seasonal changes in the amount of precipitation, will be exposed to a greater variety of environments than will the inhabitants of seasonally more constant tropical climates. On the other hand, species that are confined to a single type of habitat will, in any given climate, achieve a satisfactory degree of adaptedness more easily than will more widespread species having access to diverse habitats. The genetic structure of populations, which influences the kind and the amount of evolution that may occur, should be influenced, then, by the ecological and geographical conditions in which these populations live.

Considerations of this kind led to the formulation of a program of comparative studies of the genetic structure of species living in temperate and in tropical climates (see Year Book No. 43 [1943-1944]). This program was initiated in 1943, when the writer was able to work for about six months in Brazil, and to make, in co-operation with Dr. C. Pavan, a preliminary survey of *Drosophila* species found in the southern part of that country. Two of these species, *D. willistoni* and *D. prosaltans*, were then chosen as promising materials for intensive genetic study. Both species are inhabitants primarily of the tropical zone of the western hemisphere; their known distribution areas extend from Mexico (from southern Florida in the case of *D. willistoni*) at least as far as southern Brazil. The species *D. willistoni* is very common and widespread; *D. prosaltans* is less common and apparently more restricted ecologically.

From 1943 to 1948, the laborious but necessary work of obtaining mutant genes, making linkage maps of the chromosomes, and inducing crossover suppressors in chromosomes marked with suitable mutant genes was carried out. The genetic work on *D. prosaltans* has been chiefly in the hands of Mr. Boris Spassky and Mr. Stanley Zimmering. The cytology of *D. prosaltans* has been studied by Professor A. L. Cavalcanti, of the Universidade do Brasil, working at Columbia University. Mr. B. Spassky is almost entirely responsible for the genetical, and the writer for the cytological, aspects of the work on *D. willistoni*. The results obtained by these workers will, it is hoped, be published in special journals. What is important for the work program planned for the coming years is that the genetics of *D. prosaltans* and *D. willistoni* is now under control to the extent that analysis of the principal variables of population dynamics can be undertaken. Genetic tools are available which should permit determination of the concentrations of lethal, semilethal, and other recessive genes that may be carried in a concealed condition in natural populations; of the spontaneous and induced mutation rates; and of the frequencies of alleles among mutants found in nature or in the laboratory. Familiarity with the "maps" of the salivary-gland chromosomes should enable one to analyze the incidence of inversions that produce a variety of chromosome types in natural populations of the species named above. Some orientation tests of this kind have already been made, using samples of natural populations of *D. willistoni* collected in Brazil and sent to the writer by Dr. C. Pavan, of the University of São Paulo.

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SPECIAL PROJECTS: BIOLOGICAL SCIENCES

ROSS G. HARRISON, Osborn Zoological Laboratory, Yale University, New Haven, Connecticut. *Studies in experimental embryology*. (For previous report see Year Book No. 46.)

As in previous years, the grant from the Carnegie Institution has been applied toward the salary of a research assistant.

The year has been spent in getting ready for final publication the results of an extensive group of experiments bearing on the development of the amphibian limb, particularly on the respective roles played by the several germ layers. This has required the review of a large amount of experimental material, including preparation and study of sections and working out of the statistical significance of many of the results. In addition, further experiments have been done on the development of the internal ear, particularly on the possible influence of the mesoderm on the polarization of the auditory placode, out of which the labyrinth develops.

The main part of the manuscript on the limb has been completed. There remains to be done principally the assembling of the illustrations and the compilation of the extensive bibliography. A summary of this work follows:

The origin, development, and growth of the fore limb in Amblystoma with particular reference to the parts played by the ectoderm and the mesoderm. The paired limbs of vertebrates develop as thickenings of the mesoderm of the body wall covered by ectoderm. It is desired to ascertain the role played by each of these constituents in the development and growth of these appendages.

The methods used have been those of transplantation and extirpation. The material consisted of embryos of five species of the salamander *Amblystoma*, and of

Triturus torosus. The operations were done mostly in tail-bud stages. Homoplastic grafts were made exclusively with material from *A. punctatum*, but the heteroplastic grafts, combining tissues from two different species, which are interesting because of marked differences between certain of the species in growth rate and ultimate size, were made with nearly all the possible species combinations.

Previous investigation had shown that the area out of which the limb develops is circumscribed, being about three and a half somites in diameter and centering ventral to the fourth myotome. As far as the evidence went, the essential part of the limb rudiment was shown to be its mesodermal component, the ectodermal covering being indifferent as to place of origin (see below for exception in the case of head ectoderm). Furthermore, the antero-posterior axis of the rudiment, but not its dorsoventral axis, was shown to be polarized. No evidence was obtained to indicate that limbs could develop out of any other tissue than that of the above-described disk. Later studies by Balinsky and others, however, showed that certain structures such as ear vesicles, nasal placodes, and even nonliving material, when implanted on the flank of embryos at some distance from the limb region, would, in a small proportion of cases, lead to the induction of limbs. G. Hertwig, moreover, after grafting haploid limb buds to the flank showed by measuring the size of the nuclei that host tissues take part in the formation of the limb which develops at the seat of the graft. Though these find-

ings seem to be beyond suspicion of error, it is difficult to reconcile them in a consistent story. This has been the main incentive to the further study of the question.

By grafting whole limb buds or either layer alone, by placing tissues from other parts of the embryo in the location of the limb, by varying the orientation of the graft, and by interchanging grafts between different species, much further evidence has been obtained, showing that the mesoderm is the dominant tissue in the development of the limb and that the ectoderm plays but a subsidiary role.

For the initiation of development the limb mesoderm is essential. Normal limbs arise when this is covered by ectoderm from any part of the body before closure of the neural folds. Grafted after closure, head ectoderm suppresses development of the limb, but ectoderm from any other part of the trunk has no inhibiting effect.

Grafts of limb mesoderm alone follow the same rules of symmetry and reduplication as grafts of whole limb buds, the results depending upon their orientation on the body of the host. Grafts of limb ectoderm alone, when inversely oriented, produced reduplications in 9 cases out of 53, whereas, when normally oriented grafts were used, only a single reduplicated digit out of 50 cases was found. This difference is statistically significant, with p only slightly greater than 0.01, indicating that the orientation of the ectoderm has a minor effect on symmetry. However, the mode of reduplication is fundamentally different from that found after inversion of both layers together, since it does not involve the reversal of the primary limb bud.

Grafted limb buds from other species develop at approximately the rate characteristic of the donor. This finding is based upon observations on 26 species

permutations. Grafts of mesoderm alone behave in the same manner, showing only slight differences, of no statistical significance, from the grafts of the whole limb. Grafts of limb ectoderm alone, however, in respect to some measurements do show small differences from the normal which are statistically significant, indicating a minor effect of the ectoderm on the growth rates and ultimate size of the limb. Heteroplastic grafts, especially those between *A. punctatum* and *A. tigrinum*, reveal two other characters on which the ectoderm has a slight effect: the precocious early development of the fore limb in the former species, and the form of the digits and the interdigital web. Though these two characters are strongly dominated by the mesoderm, minor influences of the ectoderm, statistically significant, can be detected.

Further confirmation of Balinsky's heterogenetic heterotopic induction of limbs has been obtained by means of engrafted nasal placodes, though a short series of experiments with implanted ear vesicles resulted negatively.

The question of the participation of host tissues in the composition of the grafted limbs has been further studied in heteroplastic combinations. In the case of grafts in normal position, 202 cases include 195 limbs that are wholly graft specific and only 7 that give evidence of the admixture of some host material, barring certain cases in which host ectoderm is drawn out over a portion of the free limb. In grafts to the flank (5 cases), none show host influence. In grafts of mesoderm alone to the flank (19 cases), only 1 gives evidence of material from the host. In experiments like the above the odds are, therefore, about 28 to 1 against host tissues' taking part in the development of the limb.

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Cambridge, Massachusetts

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The work of the Division during the past year has to a considerable extent had direct bearing on aspects of Middle American research yet to be performed. Consequently, although the progress and results of current investigations are customarily the subject matter of the Year Book, something should be said in this report concerning plans for the future as they have taken shape during the two years since Year Book No. 45 (1945-1946, pp. 195-196) set out the desirability of review and analysis. Interruption of the normal activities of the Division by the war, conclusion of certain nonarchaeological elements of the Maya program, as well as approaching retirements made it opportune to assess past efforts, restudy the aims and methods of research, and consider most effective orientation for the Division's future effort in that field. General staff conferences consolidated divisional thinking, which benefited greatly also from informal discussion with representatives of other institutions engaged in studies of the Maya and related pre-Columbian civilizations.

For many years the Division has concerned itself with Maya history from the earliest times and over the entire area occupied by Maya-speaking peoples. Much attention has also been given to the conquest and colonial periods, to the life and customs of the present-day Maya, and to environmental conditions in the Maya country, studies designed to aid in the interpretation of the archaeological findings.

The review of what has been done to date in archaeology and allied fields has indicated that although many interesting problems remain unsolved and many

regions are still inadequately explored, a general outline of the Maya's career has been obtained. It was also indicated that indefinite continuance along the lines formerly pursued might lead to overconcentration on detail and narrowing of outlook. There seemed to be danger that, in attempting to go farther with a program covering so long a span of time and so large an area, the interests of individual workers might become so channeled and specialized that the manifest advantages of team work might be lost. For these reasons and also because of the greatly increased cost of field work, it appeared that there should be a quantitative reduction in objectives and a shift from a general study of the Maya past to an attack upon some specific phase thereof. This, it was thought, should be a period already shown to be particularly significant; one on which the attention of the entire staff could profitably be focused; and one promising to yield, within a predictable time, results of both historical and methodological importance. These considerations led to the recommendation, for inauguration in 1951, of an intensive study of the protohistoric period of northern Yucatan.

The four or five centuries preceding the Spanish conquest of Yucatan were times of stress and change. The great southern cities had been abandoned. Maya culture, however, had persisted in the north. But whereas the Maya of classic days had apparently been free to develop their remarkable civilization without foreign interference, their Yucatecan successors became increasingly affected by influences

and by actual migrations from highland Mexico and the Gulf coast. New artistic, religious, and governmental ideas surely made themselves felt. But as these are as yet only imperfectly understood, we do not know to what degree they were instrumental in bringing about the political alliances, the intergroup rivalries, and, finally, the open conflicts which seem to have caused the general decadence of the sixteenth century.

For attack on this interesting and, from the point of view of historical theory, very important period we fortunately have a large amount of information regarding preconquest times in the form of chronicles, some written by native scholars, some, at second hand, by Spaniards. The checking of these accounts is one of the most promising aspects of the proposed research. Certain students have taken them at face value; others are of the opinion that although the events recorded doubtless took place, their sequence may have been seriously garbled. The principal points at issue concern the times of founding, length of occupancy, and dates of abandonment of various cities whose ruins are identifiable. As the problems have to do with relative chronology, they can, in most cases at least, be settled by excavations at the sites in question. It is most desirable that this be done, not only to permit accurate reconstruction of what actually took place in Yucatan, but also in order that methods may be worked out for testing, by archaeological evidence, the reliability of these and of similar traditional narratives bearing on the history of other New World civilizations in Mexico and the Andean region.

The activities of the past year were a start toward clearing the decks for the proposed new program. This entails completion of all current field investigations and the making of certain surveys needed

to round out the former general study and to determine the most favorable sites for intensive investigation. The major task of most staff members, however, has been, and for the next two years will continue to be, the writing and the seeing through the press of final reports on all work accomplished under the old program.

The most important field operation of the past season was a second expedition to Bonampak, a Maya city of the Classic Period recently discovered in the densely jungled country west of the Usumacinta River in the Mexican state of Chiapas. As reported in the last Year Book, one of the temples at Bonampak contains mural paintings, remarkable for elaborateness and skill of execution, for variety and interest of subject matter, and for the splendid state of their preservation. The recording of these unique paintings has been a joint undertaking of the Institute of Anthropology and History of the Mexican government and Carnegie Institution, most generously financed by the United Fruit Company. In 1947 the murals of the first of the temple's three chambers were copied in water color and photographed with panchromatic and infrared film. The 1948 party consisted of Mr. Gustav Strömsvik, archaeologist, and Sr. Antonio Tejeda F., artist, both of the Division's staff, and Srs. Agustin Villagra and Hipolito Sanchez, artists of the Institute of Anthropology and History. Working long hours under most difficult and trying conditions, the artists succeeded, before the onset of the rains made withdrawal from the bush necessary, in completing the copies of the murals in the second and third rooms. An account of the work and a description of the newly recorded paintings are contained in Mr. Strömsvik's and Sr. Tejeda's reports below.

This second and final Bonampak expedition was the only major piece of field

work that had been planned for the winter of 1948. But as so often happens in archaeology, an accidental discovery necessitated an entirely unforeseen, and in this case most profitable, excavation.

A building boom at present going on in Guatemala City has called for the manufacture of great quantities of adobes, tiles, and bricks. The best material for these is clay which has already been mixed, tempered, and puddled, a requirement perfectly fulfilled by the fill of ancient earth mounds. As a result, a number of brickworks have been set up at the great archaeological site of Kaminaljuyu in the outskirts of the city, and many mounds are being destroyed. At the largest of all the Kaminaljuyu mounds, the workmen of a government brickworks cut into a tomb. When this was brought to the attention of the superintendent, the digging was stopped, the Division's Guatemala office was notified, and Mr. E. M. Shook made a preliminary examination. He found that the tomb lay in the center of the mound, about 13 m. above ground level and about 7 m. down from the summit. The chamber, filled with earth following the ancient collapse of its log roof, had been broken into on one side; the fragments of many vessels were scattered below. The pottery was of the Miraflores phase, confirming an earlier suspicion that, in spite of its great size, the mound had been erected during that very early period.

As the only Miraflores tomb previously known had been opened by an amateur, and as the present one was obviously very rich, Mr. Shook dropped all other work and undertook its excavation, a long and difficult piece of work because of the necessity of removing a tremendous overburden and because of the masses of crushed vessels it was found to contain. Of these there were probably over 300 (an exact count awaits their restoration).

There were also very beautiful small stone and jade containers, little stone mortars in the form of frogs, a large eccentric flint, and a so-called "mushroom stone." Neither of the latter types had been thought to have been produced at so early a date.

Important finds did not end with the tomb, for as the cutting by the government's workmen progressed, it was seen that the mound had grown by accretion and that it contains well preserved remains of a sequence of several adobe structures, the tomb having been dug into the summit platform of the next to latest. It is quite possible that others will be found below, and certain that much valuable information as to Miraflores ceremonial architecture, about which we have hitherto known nothing, will be gained as the work progresses. We are greatly indebted to the officials of the Department of Public Works, who have co-operated to the fullest extent by supplying workmen at no expense to us and by sparing uncovered structures until they can be carefully cleared, studied, and recorded. Thus we shall have opportunity to gather data on a most instructive and perhaps unique architectural complex, and one whose excavation would have been far too costly for us to undertake. Mr. Shook's report on the mound appears below.

During the season, data have also accrued as to the representation at Kaminaljuyu and vicinity of the Las Charcas and Sacatepequez phases, now proved to be even older than Miraflores. To date these are known only from their pottery and a single Sacatepequez grave found by Mr. Shook in a road-cut west of Guatemala City. Ceramically, both are as highly developed as Miraflores. The more primitive phases which must have preceded them still elude discovery.

So much for field work. Most members

of the staff have been engaged in laboratory and library studies and in writing reports, activities necessary for bringing the old program to completion and preparing for the Yucatan project.

Dr. Pollock, who is carrying the principal load of the internal administration of the Division, has also continued work on his report on the architecture of the Puuc cities of Yucatan, which contain some of the finest and best-preserved buildings ever erected by the Maya. They are believed by some students to have been products of a renaissance that took place after the abandonment of the Classic southern centers, by others to date from the latter part of the Classic Period. Solution of this very important problem is one of the objectives of the proposed new program. During the winter, Dr. Pollock visited Yucatan to check certain earlier observations on the ruins and to examine sites which are under consideration for future study. His report appears below.

Mr. Karl Ruppert, who was in charge of the expedition to Bonampak in 1947, has written a report on the architecture of that site. Further observations on some of the buildings, made in 1948 by Mr. Strömsvik, have been incorporated. The greater part of Mr. Ruppert's time has been devoted to study of the notes on, and the photographs and plans of, buildings excavated in past years at Chichen Itza that have not been published. The resultant report will complete the record of the Division's work at Chichen Itza and will form one of the bases for continued research on the architecture of northern Yucatan.

Mr. A. L. Smith has devoted the period under review to materials gathered during his survey of archaeological sites in the Guatemala highlands and in his excavation of the very rich tombs discovered by him at Nebaj. He has completed the final

report on the Division's work at Uaxactun.

Mr. R. E. Smith, who makes his residence in Guatemala and is in charge of the Division office there, has given much time to the new Guatemala National Museum of Archaeology and Ethnology. As chairman of the National Institute of Anthropology and History's committee on the museum, he has been active in the planning of the museum and, with Dr. Paul H. Nesbitt, in the installation of its magnificent Maya collections. Dr. Nesbitt's invaluable services were, through the efforts of Mr. Smith, made available by the Rockefeller Foundation. Mr. Smith's monograph on the pottery of Uaxactun is approaching completion.

Miss Tatiana Proskouriakoff has continued work on Maya sculptural art with special attention to the stelae of the Classic Period. The methods employed in her analytical study of these monuments were described in the last Year Book. It has the double aim of tracing the development of Maya stone carving, an art in which that people led all others in the pre-Columbian New World, and of determining objective criteria which will permit the dating of stelae and other sculptures that bear no inscriptions or inscriptions that have been so damaged as to resist decipherment. Miss Proskouriakoff has also made a series of restoration drawings, similar to those in her *Album of Maya architecture*, of sites and buildings in the Guatemala highlands. The necessary architectural data were gathered by A. L. Smith; field sketches of the often very spectacular setting of the groups were made by Miss Proskouriakoff.

The late Dr. Sylvanus G. Morley spent the season from November 1947 to May 1948 in Yucatan carrying forward his epigraphic researches. Although Dr. Morley's death in Santa Fe on September 2, 1948, occurred after the close of the period

covered by this report, there must here be voiced the deep personal grief of his colleagues of the Division, and their sense of the irreparable loss to Maya research in the passing of its most devoted and most eminent student. A statement regarding his great contribution to knowledge of the Maya will be prepared for the next Year Book.

Mr. Thompson has continued his studies of Maya hieroglyphic writing. The introductory volume to this work is virtually completed, and it is anticipated that the book will be ready for the publishers next year. Mr. Thompson also has given his time as editor of the Division's series of Notes on Middle American Archaeology and Ethnology.

The Chairman spent the winter in Guatemala working on the collections of potsherds made by A. L. Smith during his highland survey. He also made notes on, and photographed the pottery and artifacts from, the Nebaj tombs for a joint paper with Mr. Smith. The Nebaj materials are of much interest, not only for the fine preservation of the pottery and the outstanding beauty of the jade carvings, but also because both pottery and jades evidence active trade relations between Nebaj and Classic centers in the lowlands. The Chairman has continued to act as archaeological adviser to the United Fruit Company, which is engaged in the excavation and restoration of the important ruins of Zaculeu in western Guatemala.

In the field of history and native Maya literature, Mr. R. L. Roys spent a considerable part of the year in proofreading and indexing the monograph on the Chontal Indians of Acalan-Tixchel by himself and Dr. F. V. Scholes, now in press. He also made an intensive study of Maya prophecies to investigate the meaning of various obscure phrases and mythological references. For this purpose he

was obliged to make a further study of the Ritual of the Bacabs, a book of medical incantations. The results of his research may form an extensive appendix to Mr. J. E. S. Thompson's work on the Maya glyphs, as it is possible that certain of the stock phrases of the prophecies and incantations also formed parts of the hieroglyphic inscriptions and that cross-identification may throw light on the meaning of glyph groups not now decipherable. Mr. Roys has been made Honorary Research Professor of Anthropology at the University of Washington.

Dr. Scholes, now Dean of the Graduate School and Vice-President of the University of New Mexico, continues, for the Division, his research and writing on the history of Yucatan, assisted by Miss Eleanor B. Adams. In addition to work on the above-mentioned joint monograph with Mr. Roys on Acalan-Tixchel, he has been engaged in several studies preliminary to, and laying foundations for, his *Yucatan in the sixteenth century*, which will form the fourth and final volume of a series on the early history of the peninsula, the first being Mr. Roys' *Indian background of colonial Yucatan*, already published; the second and third, Dr. Scholes' and Mr. Roys' *The Maya Chontal Indians of Acalan-Tixchel*, and Dr. Robert S. Chamberlain's *Conquest of Yucatan*, both in press.

Statements on the work of Miss Anna O. Shepard on ceramic studies, of Dr. E. H. Morris on Southwestern archaeology, of Dr. Norman McQuown on Maya linguistics, and of Mrs. W. H. Harrison on Division publications form part of the body of this report.

BONAMPAK EXPEDITION

GUSTAV STRÖMSVIK

A report on the 1947 expedition to the ruins of Bonampak, under the direction of

Mr. Karl Ruppert, of Carnegie Institution, financed by the United Fruit Company, and participated in by the National Institute of Anthropology and History of Mexico, appears in Year Book No. 46 (1946-1947, pp. 177-179). The primary objective of that expedition was to copy the extraordinary mural paintings discovered there by Mr. Giles G. Healey in 1946. Owing to various delays and transportation difficulties, the murals in only one of three decorated rooms were completely copied, but these proved to be so magnificent and so important that the United Fruit Company continued its support in 1948, thus making it possible to finish copying the paintings and to obtain additional data on the site. The 1948 expedition was organized by Sr. Antonio Tejeda F., artist, and Mr. Gustav Strömsvik, archaeologist, both of Carnegie Institution. Sr. Agustin Villagra, of the National Institute of Anthropology and History, again participated, and, to insure completion of the work, the services of Sr. Hipolito Sanchez, of the Archaeological Museum in Campeche, were obtained.

The earlier expedition had encountered great difficulties in procuring mule transportation to the ruins; this year, with chicle exploitation in the region stopped, mules were virtually unobtainable. In view of the possibility that equipment and supplies might have to be transported on the backs of men, impedimenta were cut down to an absolute minimum, in both weight and volume.

Equipment and supplies were assembled in Merida and shipped by rail to Tenosique. On March 1, 1948, Tejeda joined Strömsvik in Merida, and next day they proceeded by train to Campeche, where they were met by Villagra and Sanchez. Ways and means for reaching the ruins were discussed. Four men were hired to carry the packs from Santa Clara chicle

station to the ruins, should that prove necessary. Santa Clara was chosen as point of entry because it is the landing field nearest the ruins and the trail is fairly good.

On March 5 the expedition took the train from Campeche to Tenosique, there being no regular air service over that route. At Tenosique, through connections and friendships made last season, twelve mules and two muleteers fortunately were obtained and sent overland, via Piedras Negras, to Santa Clara. On March 12 a bi-motored plane was chartered at Tenosique to fly the staff and equipment to Santa Clara; in the evening of the same day the mules and boys arrived at the chicle station. After a day's rest, the expedition got under way into the jungle, the men taking turns walking and riding. The trail was much overgrown and very difficult to follow, and several times it was lost. In spite of these delays, the ruins were reached in two and a half days without serious incidents.

On the trail to the ruins, numerous mounds were noticed, but no cut stones or sculpture. Such remains occur some 300 m. south of the chicle camp at Santa Clara, by the Lacanha River, and at an abandoned chicle camp called Moguel, about halfway between the Lacanha and Bonampak.

At the ruins, the area in front of Building 1, which had been cleared the previous year, was completely covered by lush vegetation, but the thatch house built last season would still give shelter. The artists immediately set to work at preparations for copying the remaining paintings. Getting camp settled, the supplies protected against occasional heavy showers, and camp routine established, came first. Afterward, time was spent in helping the artists and in preparing the walls for copying. Scaffolding had to be built of materials at hand; the kerosene lamps used for illumination created so much heat and carbon

monoxide in the rooms that a fan had to be constructed to force in fresh air to enable the artists to work. Owing to transportation difficulties, a very limited supply of canned goods and meats had been brought along, but the neighboring Lacandon Indians supplied some sweet potatoes, plantains, and green corn. One of our workmen turned out to be a good hunter, and with our shotgun supplied the necessary meats.

When time permitted, notes were made of further details to complete the Bonampak map. Numerous excavations were made in courts and by stairways to find ceramic specimens, but in vain. During the interval since last season, the major part of the complicated roof ornamentation of Building 4 had fallen, but all details are preserved in Ruppert's careful architectural notes of the site.

The work of copying the paintings in the remaining two rooms of Building 1 progressed according to a carefully calculated program and was completed during the six weeks allotted for the task. According to agreement made beforehand, the expedition was met by a plane at the Santa Clara field April 24. The expedition had completed its task.

BONAMPAK EXPEDITION

ANTONIO TEJEDA F.

Until the discovery of the Bonampak murals, nothing had led us to suspect the great technical ability and the extraordinary mastery of drawing and of composition possessed by the Maya painters of the Classic Period.

The method used in producing the murals was apparently as follows. On the still moist plaster the artist drew the entire composition in a sepia color, which in some instances is blood red, perhaps owing to the action of time. There is no doubt that

the design outline and major color backgrounds were painted directly on the fresh wall surface, because fallen pieces of the paintings show the pigment deeply embedded in the plaster. The relatively large compositions, covering an entire wall surface, were presumably accomplished in this fresco technique by laying the final coat of plaster on one section of wall at a time, and possibly by the use of assistants to apply color after the composition had been rapidly outlined. In some parts of the compositions corrections may clearly be observed, left unchanged by the artist rather than risk damaging the wall. In order to intensify color for shading, to obtain different tones, or to strengthen and bring out details, the artist used pigments mixed with some unknown adhesive, which did not materially affect the permanence of the colors. The superposition of colors in certain areas is revealed by the fallen pieces referred to above. The entire technique, fresco-tempera, is curiously like that of the Italian Renaissance. Only mineral paints seem to have been employed. Their selection and preparation were astonishingly skillful, as they have retained their tone and brilliance through the centuries, in spite of the most adverse climatic conditions.

The paintings of Rooms 2 and 3 have now been faithfully copied; those of Room 1 were copied in 1947. Those of each chamber are independent in regard to composition and descriptive action of the scenes, a unit of arrangement and subject matter, but all three paintings form a most harmonious total composition. Each must be studied by itself, however, in order to appreciate the resourcefulness with which its elements have been fitted to the available spaces, and the skill with which individual figures and groups have been balanced.

The paintings of Room 2 represent the

highest plane of artistic conception. On one wall there is depicted a stirring scene of battle. Another part of the room shows the torture of prisoners. A very high place in the history of art must be granted to these renderings, which, for balance, harmony, dynamic action, and the expression of ferocity, triumph, suffering have seldom been surpassed. One figure, that of a sacrificed captive, exhibits a knowledge of foreshortening that is truly marvelous.

In Room 3, as in Room 1, are depicted scenes of ceremony, including winged figures with rich and elaborate headdresses. As spectators there are several priests wearing long white robes similar to those of persons shown in Room 1. There are also several women seated on a dais, at whose base is another woman holding a child in her arms. These women and the priests form groups of great interest and charm.

Many of the glyphs found in Rooms 2 and 3 have suffered severely, and their decipherment will be difficult. It seems that the artists of Bonampak were more concerned with the depiction of events, with action, and with the magnificent costumes of individuals than with recording of chronological data.

The paintings in Room 1, copied last year, remain in excellent condition, and the cleaning of the walls permits them to be seen more clearly. Those of Rooms 2 and 3 are less well preserved because of the heavy calcareous coating deposited on their surfaces, the filtering of water through the walls, and the stains produced by fungi and bat droppings.

During the stay at Bonampak, drawings were made of the sculptured lintels of the doorways opening into the three painted rooms. These are in low relief, each depicting a warrior with a prisoner. They had once been painted in the same polychrome style as the murals. Only a few

patches of the colors are left, but these may make possible color restorations of the carvings.

The National Institute of Anthropology and History faces a very difficult problem in the preservation of the wonderful paintings of Bonampak. Some sections of the stucco background, which has an approximate thickness of 1 cm., have loosened from the walls because of the humidity. The open spaces thus formed collect moisture and are used as nesting places by insects. This is causing parts of the murals to fall, and such areas require prompt attention. Attempt was made by the 1948 expedition to clean the building as much as possible. All vegetation growing on the structure was removed in order to halt the destruction caused by roots, many of which have penetrated the walls and produced cracks and passages which allow water to enter during the downpours of the rainy season.

GUATEMALA HIGHLANDS

EDWIN M. SHOOK

Two of the most significant archaeological discoveries of the past year in Guatemala were made accidentally. The first was in Kaminaljuyu on the outskirts of Guatemala City, and the second, 29.5 km. west of the city near Santa Maria Cauque, in the Department of Sacatepequez.

The latter find was made en route to Tiquisate by Mr. Shook and his assistant, Mr. Henry Morgan, as they were passing through a new road-cut for the Roosevelt Highway, where it skirts the foothills along the west side of the Rio Sacatepequez valley. A low spur of the mountains had been cut to a depth of 4 m. below the original surface with heavy road equipment. Prior to the excavation, no artificial mounds, pottery fragments, or other remains had appeared on the surface to

indicate that a town site once existed there or in the immediate vicinity. About 0.75 to 1.0 m. below the surface, however, a number of ancient pits and graves were encountered. The pits, similar to ones frequently found in Kaminaljuyu and Salcaja, were oval in profile, varied from 2.0 to 3.0 m. in depth, and were filled with ancient rubbish. Many graves were disturbed by the heavy machinery, as was evident from the number of freshly broken pottery vessels, human bones, and jade beads scattered along the roadbed. One grave, left partly exposed in section, was excavated by Shook and Morgan. A vertical shaft had been sunk to a depth of 2.5 m. and an adult buried at full length on its floor. Surrounding the body were thin stone slabs set on edge. The left side of the skeleton, the slabs, and whatever mortuary furniture existed on that side had been swept away by the bulldozers. The remainder of the skeleton was *in situ* with eight pottery vessels aligned at the right of the body, several obsidian lancets, and a single jade bead. In addition to the grave contents, a large sample of pottery and artifacts was collected from the road-cut. All this material proved to be Pre-Classic and to be intermediate between Las Charcas, the oldest phase at present identifiable in the highlands of Guatemala, and the Miraflores phase of Kaminaljuyu. It has been given the name Sacatepequez from the locality in which it was found; its chronological position between Las Charcas and Miraflores was ascertained by the excavations at Kaminaljuyu described below.

The diagnostic pottery of the Sacatepequez phase is a white ware with medium-textured white paste and a well smoothed, polished surface. Vessel exteriors were often decorated by simple incising, grooving, or modeling, and by plain bands or broad-line geometric designs in red paint.

This white or red-on-white ware comprised 55 per cent of the Sacatepequez collection. It occurs abundantly in the Guatemala and Antigua valleys, and has been noted as far west as San Cristobal in the Department of Totonicapan. Worn fragments came from the fill of Structure E-III-3 in Kaminaljuyu, but no vessel of white or red-on-white ware was found in the richly stocked Miraflores tomb in that structure. Such typical Miraflores features as hairline and coarse incised brown-black ware, fine red ware, "Archaic"-type figurines, effigy rim-heads, and secondary vessel supports were entirely lacking from the Sacatepequez material. Fragments of metates, manos, and comales indicated that even at this early date the people were sedentary and had developed a secure economy based on the cultivation of corn.

The destruction of the great archaeological site of Kaminaljuyu on the southwest edge of Guatemala City has accelerated since 1940 owing to the encroachment of the modern city in that direction and to the heavy demand for bricks, tiles, and adobes. The material for these is derived largely from the ancient mounds. Many groups and individual mounds have been removed completely since the Institution surveyed Kaminaljuyu in 1936 and 1939. Recently, the Department of Public Works of Guatemala acquired the largest single mound, Structure E-III-3, for the manufacture of bricks. Last November, after most of the eastern half of E-III-3 had been cut away, the workmen, in making another vertical cut downward from the top of the mound, encountered human bones; pottery and stone vessels, some containing mineral paints; several jade ornaments; and a carved "mushroom" stone. Fortunately, the superintendent of the brickworks, Sr. Moises de Leon, halted the work and reported the finds to the Director of Public Works. The Director in turn advised

the Minister of Public Education and the National Institute of Anthropology and Ethnology. Mr. Shook was requested to investigate. The above-mentioned objects proved to have come from a tomb, the most richly stocked to be unearthed to date in Guatemala, and, of more importance, the first major tomb to be found dating from the Miraflores phase of the Pre-Classic Period. Excavations were undertaken with labor generously supplied by the Department of Public Works, under the combined direction of Dr. Kidder and Mr. Shook, ably assisted by Sr. Gustavo Espinoza.

The tomb had been cut into the top of an existing pyramid, 13.40 m. above ground level, in a series of three terraces stepping in on all four sides toward the burial chamber, a rectangular pit measuring 3.20 m. north-south and 1.70 m. east-west. Four large wooden posts had been set, one in each corner, to support two heavy north-south longitudinal timbers, on which were placed close together the east-west transverse beams that roofed the chamber. Prior to roofing, the burial of a most important personage took place, the body apparently laid on the floor. The position of the skeleton could not be determined because the eastern half of the floor had been cut away by the brick-factory workmen. However, the jade ornaments which must have accompanied the body were found near the center. Abundant pottery vessels and other offerings had been piled to the height of the roof around the tomb's walls. Further mortuary offerings were heaped on the roof and the terraces. The remainder of the pit was filled with mixed earth to the level of the pyramid top, and a thin adobe floor laid over the tomb area. Some slumpage occurred from settling, and perhaps from the breakage of the vessels by the pressure of the fill, and several other floors were added to level

up. Also a small rectangular adobe platform, perhaps a shrine, was built directly above the tomb. It appears that these floors and the platform were temporary measures to permit the continuation of ceremonies dedicated to the deceased, while a completely new pyramid was being constructed to encase the one into which the tomb had been cut.

The tomb's location in the principal Miraflores structure at Kaminaljuyu, the enormous work involved in its construction, and its rich furnishings give ample evidence of the position of great honor and esteem held by the deceased. The mortuary offerings included some 300 pottery vessels; finely wrought vessels of marble, greenstone, and jade; stone mortars and pestles, several of the mortars carved in the form of toads; the carved "mushroom" stone previously mentioned; about 100 obsidian-flake lancets; one large eccentric flint; jade beads, earplugs, and pendants; shell ornaments; mica sheets; mineral paints; and objects made from armadillo shell. Perishable items were evidenced by textile, mat, and wood impressions. Among the pottery vessels were effigies, pieces decorated with painted stucco, and at least eight with covers, a feature common for certain forms in the later Esperanza phase but heretofore unknown in the Miraflores phase.

While the tomb excavation proceeded, careful watch was maintained on the digging done by the brickworkers on other parts of the mound. We were able to determine that E-III-3 consisted of a succession of pyramidal adobe structures, facing south, built one over the other. Each adobe accretion had an elaborate frontal platform which underwent several renovations before being buried by the next pyramid. Low, rectangular platforms with postholes at each corner were found on one frontal platform; on the floor near

by, severely burned rectangular depressions containing ashes and charcoal. The latter appeared to be hearths for ceremonial fires, the former to support small individual shrines of perishable material.

The entire building activity in Structure E-III-3 apparently took place during the Miraflores phase. No evidence was seen in the fill of the latest pyramidal addition or on its surface that the mound had been used during the subsequent Classic and Post-Classic Periods.

Other activities of 1947-1948 were as follows:

The southwestern section of Kaminaljuyu was mapped, completing the surveyed plan of the site. The principal group there consists of eight mounds arranged in an orderly manner on the four sides of a plaza. The court and area surrounding the mounds had been excavated in ancient times to a depth of 4.0 m. from the original ground level. This left the eight units, each 4.0 m. high, of undisturbed sterile volcanic deposits. Five of the mounds remained unaltered thereafter, whereas the heights of three were increased in the normal manner by the addition of more material.

Several weeks were spent in Quetzaltenango recording in detail the archaeological material recovered by Sr. Vitalino Robles from the ruins of El Paraiso (Year Book No. 46 [1946-1947], pp. 182-183). This completes the field data for the publication on the ruins of El Paraiso.

One month was devoted to the study and photographing of the various private collections in Tiquisate. Most of the specimens in these collections were found in the Tiquisate zone during the building of roads, the cutting of drainage canals, and the installation of pipe lines. The stratigraphic excavations made in 1947 (Year Book No. 46, pp. 183-184) permit the utilization of this large body of ma-

terial. Its full value may be appreciated when one considers that several years of intensive work and the expenditure of a large sum of money would be required to recover a comparable amount of data. The United Fruit Company again generously provided facilities and assistance in this study.

Mr. Stanley H. Boggs, after two years as archaeologist of the Zaculeu Project, resigned in the summer of 1947. Mr. Richard Woodbury was appointed in his place; by request of Dr. Kidder, Mr. Shook spent three weeks in Zaculeu helping Mr. Woodbury to become acquainted with the work accomplished during the previous seasons. At the same time this gave Mr. Shook opportunity to improve his knowledge of Zaculeu ceramics. The pottery sequence showed that Zaculeu was founded during the Early Classic Period and was continuously occupied until the conquest by the Spaniards in A.D. 1525. Only a small amount of material has been recovered representing certain periods, especially the Late Classic. This may be due more to failure to locate structures than to lack of occupation of Zaculeu during those times.

A brief reconnaissance was made in El Salvador in the company of Mr. Boggs. The major archaeological zones of Campana San Andres, Chalchuapa, and Cihuatán were visited, as well as many smaller sites. Cihuatán proved to be a large site strategically located on a slight elevation overlooking the broad, fertile valley of the Acelhuate River, a tributary of the Lempa. House platforms, temple pyramids, a late-type ball court, and the wall enclosing the central plaza are well preserved. The house platforms are concentrated on the gentle slopes outside the walled area, suggesting the typical Latin American town of today, where the people live around a religious and civic administrative nucleus.

Cihuatán appears to have been an important center in the Post-Classic Period and may have been occupied at the time of the conquest.

A short trip to Tikal with Dr. and Mrs. E. Wyllys Andrews was made to estimate the time and equipment necessary to obtain a record of the architectural remains readily available there without excavation. There exists in Tikal more exposed and well preserved architecture than in any other site known in the Department of Peten. It would require one field season to photograph and study adequately this material without undertaking more than minor excavations.

ARCHITECTURAL SURVEY OF YUCATAN

H. E. D. POLLOCK

It was noted in a previous report on the Architectural Survey (Year Book No. 45 [1945-1946], pp. 206-207) that the immediate task in hand was the preparation of the illustrative material that will form the body of the final report on this work. In the working over of the notes, sketches, and photographs that are the raw data of the survey, it was inevitable that certain gaps, certain loose ends, should become apparent. Some of the lacunae and uncertainties thus uncovered seemed sufficiently important to merit checking in the field. Dr. Pollock consequently spent March and part of April 1948 in Yucatan.

As a good proportion of the checking concerned the ruins of Uxmal, this site was used as a base. The work was greatly facilitated by the kindness of Sr. Jose A. Erosa Peniche, archaeologist of the National Institute of Anthropology and History, in offering the use of the Institute's living quarters at the ruins. Approximately three weeks were spent at Uxmal, during which time trips were made to the ruins of Huntichmul I, Kiuic, and Xculoc. Dr.

Pollock was so fortunate as to have the company of Dr. Morley during a part of his stay at Uxmal.

Toward the end of March Mr. Shook joined Dr. Pollock in Merida and the two visited the well known site of Mayapan for the purpose of checking the existing map of the ruins in order to determine the advisability of additional mapping.

The small but excellent regional museum in Campeche was visited next. As the new highway connecting Merida and Campeche had recently been opened for traffic, the opportunity was taken to traverse this road, which continues the old Merida-Uxmal highway to the ruins of Kabah, thence to Bolonchen-Ticul, to Hopelchen, and westward to Campeche. A number of ruined sites were observed along the road, the most notable being that marked by a high, pyramidal structure which may be seen at a distance of 2 or 3 km. north of the highway between Uxmal and Santa Elena. Mr. Shook is of the opinion that this structure is part of a large site that he observed from the air while flying from Guatemala to Merida. It is probable that these are the ruins of Xcoch, discovered over 100 years ago by John L. Stephens and, so far as is known, never revisited. Also worthy of mention is a group of remains, immediately west of Hopelchen, that have been cut through and in part demolished by the construction of the new highway.

The work of the Architectural Survey has been retarded by the administrative duties assumed by Dr. Pollock in 1947. The results of this research are not so near publication as might have been expected at this time. Although the great task of drafting the illustrative material is virtually completed, this material and the photographs are not yet arranged in final form for publication. Only a small part of the text has been written. It is hoped that

Dr. Pollock will be able to give more of his time to this work in the ensuing twelve months.

CERAMIC TECHNOLOGY

ANNA O. SHEPARD

Plans for the use of punch cards in comparative ceramic studies were carried forward during the year. The desirability of establishing a project in co-operation with institutions active in Mesoamerican archaeology was considered, and in October 1947 a tentative outline for such a project was discussed with representatives of four other institutions. Although the idea was favorably received, it was deemed inadvisable to undertake at this time the work of establishing and bringing up to date a number of files. Our own requirements, however, afforded opportunity to initiate the system and demonstrate its value. The Division's ceramic photographic file demanded revision and is therefore being converted to punch cards. In order to do this without prejudice to the co-operative project, which it is hoped will be established later, a detailed memorandum and a draft of the proposed code were submitted to all archaeologists with whom the co-operative plan had been discussed. The code adopted permits sorting by provenience, chronology, ceramic classification, and a large number of shape and design classes and features. It is flexible and provides for expansion as our information increases. In order to manage the project efficiently, a record card has been drafted which will at once simplify and systematize recording and enable untrained clerical help to type and punch cards without supervision.

The punch-card index of petrographic sections started last year has been completely typed and largely punched. It is now being used in analytical work.

One difficulty in the conduct of a technological laboratory is to strike a proper balance between studies directed toward the solution of well defined problems and the routine analytical work which is always in demand to check classifications and to complete descriptions. The tendency of those unfamiliar with the uses and values of laboratory methods in archaeology is to think in terms of the latter alternative. Consequently a special project has been chosen, in order to acquaint archaeologists more generally with the applications and significance of ceramic technological data. It was felt that the study selected should be one which gave reasonable promise of definitive results elucidating a major problem, and that it should be conducted in co-operation with another institution. Extensive correspondence was carried on with archaeologists in the Peruvian, Mexican, and Southwestern fields, and preliminary examination was made of a number of samples in order to evaluate the proposed projects. Among the sherd lots submitted, two from Mexico were of particular interest, a small collection from the Tampico-Panuco area submitted by Dr. Gordon Ekholm, and samples from Tres Zapotes and La Venta selected by Dr. Philip Drucker. The Tampico-Panuco sherds show an exceptionally wide variety of readily distinguishable pastes which would undoubtedly aid materially in identifying intrusives and tracing the foreign contacts which appear to have affected many times the course of ceramic history in this area. But comparative material was not available, and there is no opportunity at present to follow out an investigation. The Tres Zapotes-La Venta pottery was of interest because of the prominence of fine-paste ware, varieties of which are important among the late types of Yucatan. Although a number of specific varieties could be defined, and

marked differences in composition were noted between those of La Venta and those of Tres Zapotes, this study had the same limitation as the Tampico-Panuco. Attention was therefore turned to the Pueblo field, where conditions are more favorable for following trade in pottery. There is also the advantage, in this field, of the co-operation of an archaeologist who has long been interested in ceramic technology, Dr. J. O. Brew, Director of Peabody Museum at Harvard. The problem chosen relates to the position of Mogollon culture, a subject of long and continuing controversy. An attempt will be made to test the actual contacts of this culture with those of the north through the location of the sources of the supposed Mogollon intrusives. Results should supply a sound basis for the study of the interactions of the two cultures during the early ceramic period.

At the request of Dr. Alfonso Caso, a sample of Monte Alban sherds, including ninety-one of his pottery types, was studied to learn whether or not his classification is consistent with the relationships indicated by paste composition and other technical features. The opportunity to examine this pottery was welcome because of the archaeological importance of the site and because it gave reference data for comparison of various gray sherds from Uaxactun, Copan, and other Maya sites, sherds which from time to time have been questioned as possible intrusives from Monte Alban. Since much Monte Alban pottery is smudged, thermal tests as well as petrographic analyses were made. Two principal classes of paste were found: one, orange-colored, is tempered with quartz sand; the other, cream or buff, is tempered with heavily weathered feldspathic sand. Each paste occurs in a range of types including coarse, heavy, and fine-finished wares, and likewise each includes smudged and oxidized types, from which it would

appear that these pastes mark the indigenous pottery. They correlate to a certain degree with four color groups—Yellow, Cream, Gray, and Brown—into which Dr. Caso divides his types. The majority of the Yellow types have quartz sand temper, and the exceptions are distinctive, whereas feldspar sand is not represented in the sample of Yellow. The majority of the Gray types are smudged variants of the Yellow, but in this group there is a small series of thin-walled, well finished sherds with feldspathic sand temper. The Cream ware types all have feldspathic sand temper with the exception of two fine pastes of the texture of untempered clay. The Brown group is mixed; both of the common pastes are well represented and there are a number of distinctive ones. In addition to the common pastes, distinctive pastes were found in sixteen types. In many cases these were associated with uncommon shapes, finishes, and decorative techniques. Many of them doubtless are intrusives. One of the well known intrusives present is the important and widely traded ware, thin orange. It closely resembles in color certain of the indigenous types, but identification was made certain by temper analysis. Among the unique specimens was a white-rimmed black sherd comparable in paste to pottery of this kind from Tres Zapotes and La Venta. Neither of the common Monte Alban pastes is represented in any of the Gray sherds from Maya sites so far sent to the laboratory.

The rest of Miss Shepard's time during the year was devoted to the preparation of a general ceramic book for the use of archaeologists.

EARLY CULTURES OF SOUTHWESTERN UNITED STATES

EARL H. MORRIS

Dr. Morris spent the year working toward the completion of publications in

the field of Southwestern archaeology. These reports deal with excavations, and materials recovered during the course thereof, conducted in southwestern Colorado and northeastern Arizona before the war. Time was about equally divided between actual preparation of manuscripts and describing and photographing several hundred specimens. The principal reports in progress are one on Basket Maker II dwelling sites near Durango, Colorado; one on Basket Maker III ceramics from caves in the drainage of Black Horse Creek in Arizona; and a third which will present in detail the weaves and decorations of Anasazi cord sandals over the interval from A.D. 217 to about 1250.

In April 1948 Dr. Morris gave the Annual Research Lecture at the University of Colorado. His subject was "Archaeological experiences at Quirigua, Guatemala."

LINGUISTIC RESEARCH

NORMAN A. McQUOWN

Dr. McQuown spent the time from July 15 to December 15, 1947 on a field trip to Tancanhuitz, San Luis Potosi, Mexico. The purpose of this trip was to gather material for the completion of the descriptive phase of the work on Huastec. A short field trip had previously been undertaken (November-December 1946), and the material gathered at that time was used as a basis for a phonemic analysis, which in turn made it possible critically to examine the Huastec materials collected by Andrade (see Year Book No. 46 [1946-1947], pp. 193-195). This examination made it apparent that the transcription of the recorded text materials would have to be checked and completed in those aspects (stress, vowel length, glottal stop) in which it fell short, and that further paradigmatic material (without which it would be difficult or impossible to work out the gram-

mar) would have to be obtained, working with an informant in the field.

During the four months actually spent in the field, the following was accomplished: (1) A Huastec grammar and dictionary file of approximately 10,000 entries was collected; (2) a Huastec grammar, in its essentials, was worked out; (3) the transcription and translation of the Potosi Huastec texts recorded (phonographically or phonetically in notebook) by Andrade were checked and completed; (4) a half-dozen new texts (of ethnographic or folkloristic interest) were taken down; and (5) a short vocabulary of the Veracruz dialect was obtained.

The Potosi materials alone will constitute an adequate basis for comparison with the other Maya languages. The Veracruz materials (Andrade's and McQuown's) may be worked up in the form of notes to the Potosi materials, or may be used as a basis for eventual further study, pending the collection of additional data from Veracruz.

The major part of Dr. McQuown's time from January to June 1948 was taken up with the teaching of linguistics in the Department of Anthropology at the University of Chicago, the remainder with the elaboration of the Huastec materials.

In process, and near completion, is a Spanish-Huastec dictionary file, based on the collected materials. This will be incorporated into the Spanish-keyed comparative Maya vocabulary, together with materials from other Maya languages.

Now completed, in card file and in notebook, are: (1) a descriptive grammar of Huastec; (2) a collection of forty Huastec texts, completely retranscribed and retranslated from the phonetically and phonographically recorded materials; and (3) a dictionary file (Huastec-Spanish) covering these materials. This concludes the descriptive phase of the Huastec work.

HISTORY OF SCIENCE

GEORGE SARTON

Introduction to the history of science.

Most of Dr. Sarton's time was devoted to seeing volume III through the press and to preparing the elaborate indexes—general, Greek, Chinese, Japanese. Part I of Carnegie Institution of Washington Publication 376, vol. III appeared in the fall of 1947; part II in the spring of 1948.

Editing of Isis and Osiris. During the year were published nos. 109/110 of *Isis*, forming the second half of volume 37; nos. 111–114, forming volume 38; and nos. 115/116, forming the first half of volume 39. These eight numbers of *Isis* comprise a total of 580 pages, with 7 plates and 38 figures, and include 43 main studies, 133 notes, 45 reviews, and about 2400 bibliographic items. Volume 8 of *Osiris* is being printed in Belgium.

Lecturing in Harvard University. A course of thirty-five lectures on American science was delivered.

Dr. Sarton sailed for Europe February 19, 1948.

PUBLICATIONS

MARGARET W. HARRISON

The Maya Chontal Indians of Acalan-Tixchel: a contribution to the history and ethnography of the Yucatan Peninsula (Publication 560), under the joint authorship of France V. Scholes, of the University of New Mexico, and Ralph L. Roys, is in revised page proof; the very detailed index, prepared by Mr. Roys and Miss Eleanor B. Adams, is in galley proof. It is hoped that the book will be issued before the end of 1948. The third volume in this series of historical studies, *The conquest and colonization of Yucatan, 1517–1550* (Publication 582), by Robert S. Chamberlain, is printed and will be distributed in August. Dr.

Chamberlain's manuscript *The conquest of Honduras and Higuera* awaits editing.

Plumbate: a Mesoamerican trade ware (Publication 573), by Anna O. Shepard, was issued in June. The experiment of fitting the text to face its pertinent illustration has been considered successful, and the convenience of comparison worth the slight cramping of wordage.

The preprints of volume IX of Contributions to American Anthropology and History (Publication 574) were also issued in June; the full volume will follow shortly. It contains four papers: *An archaeological reconnaissance in the Cotzumalhuapa region, Escuintla, Guatemala* (no. 44), by J. Eric S. Thompson; *Textiles of pre-Columbian Chihuahua* (no. 45), by the late Lila M. O'Neale, of the University of California, Berkeley, to which is appended a chemical analysis of the coloring matter by Michael Kasha, of the University's Department of Chemistry; *The governorship of the Adelantado Francisco de Montejo in Chiapas, 1539–1544* (no. 46), by Robert S. Chamberlain; and *The symmetry of abstract design, with specific reference to ceramic decoration* (no. 47), by Anna O. Shepard.

The first paper in volume X of the Contributions (Publication 585) is in press: *The Maya chronicles* (no. 48), by Alfredo Barrera Vasquez and Sylvanus G. Morley. The manuscript of the next two papers, *Guide to the Codex Perez* (no. 49), by Ralph L. Roys, and *The Pendleton ruin, Hidalgo County, New Mexico* (no. 50), by A. V. Kidder, H. S. Cosgrove, and C. B. Cosgrove, is ready for editing and will be presented for publication in the fall.

The artifacts of Uaxactun, Guatemala (Publication 576), by A. V. Kidder, was published in October 1947. As part of the general survey of work at Uaxactun, it will be followed by reports on the excava-

tions and pottery by A. L. and R. E. Smith, respectively. That on the excavations is ready for publication.

The addition of nine papers during the year filled the third volume of Notes on Middle American Archaeology and Ethnology. All but one, this by Dr. Kidder and listed in the bibliography at the end of this report, came from authors outside the Institution: *The codex of Tonayan* (no. 84), by R. H. Barlow; *Elements of Maya arithmetic, with particular attention to the calendar* (no. 85) and *Did the Maya have a zero?* (no. 90), by Charles C. Ful-

ton; *Cuchumatán textiles: the course of an error* (no. 82), by Oliver La Farge; *Observation of the sun among the Ixil of Guatemala* (no. 87) and *Some remarks on Maya arithmetic* (no. 88), by Richard C. E. Long; *Representations of temple buildings as decorative patterns on Teotihuacán pottery and figurines* (no. 83) and *Certain types of stamped decoration on pottery from the valley of Mexico* (no. 86), by Hasso von Winning. The Division will issue the title page, table of contents, and index for the volume in September 1948.

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SPECIAL PROJECTS: HISTORICAL RESEARCH

E. A. LOWE, The Institute for Advanced Study, Princeton, New Jersey. *Collection and study of paleographical material*. (For previous reports see Year Books Nos. 9 to 35 and 37 to 40.)

The war interrupted all field work on *Codices latini antiquiores*. As soon as it was over, however, and the manuscripts became once more accessible to students, Dr. Lowe sailed for Europe to begin work on the revision of the material for volume V, which deals with the oldest Latin manuscripts of Paris. During his half-year's stay there (from the end of April to the end of October 1947) the larger part of volume V was got ready for press, and copy was sent to the printer in October. The revision of the rest of this volume should be completed during the summer of 1948. Fortunately most of the manuscripts that concern this volume were photographed before the war; the remaining photographs, it is hoped, will be procured in the summer of 1948.

Volume IV of *C.L.A.* was published on May 15, 1947, with the imprint "At the Clarendon Press, Oxford." It contains more facsimiles than any of the preceding volumes, and its introductory matter is more extensive.

Grateful mention must be made of the extraordinary facilities which Dr. Lowe and his assistants have been enjoying at the Bibliothèque Nationale of Paris, thanks entirely to the liberal attitude and generous interest of M. Porcher, Keeper of the Manuscripts. It is also gratifying to be able to report that contact has been re-established with certain scholars abroad whose assistance has been of signal value to the publication of *Codices latini antiquiores* in the past.

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